PCT WORLD INTELLECTUAL PROPERTY ORGANIZATION
International Bureau

INTERNATIONAL APPLICATION PUBLISHED UNDER THE PATENT COOPERATION TREATY (PCT)

(51) International Patent Classification 7 :
A61K 31/415, 31/445, 31/40, 31/405, C07D 231/12, 231/14, 209/14, 209/18, 207/32, 207/40, 209/42, A61P 31/04, C07D 401/04, 231/56, C07C 233/54, C07D 471/04, 249/10 // (C07D 471/04, 249:00, 221:00)

A1

(11) International Publication Number: **WO 00/37074**

(43) International Publication Date: 29 June 2000 (29.06.00)

(21) International Application Number: PCT/EP99/09577

(22) International Filing Date: 7 December 1999 (07.12.99)

(30) Priority Data:
9828005.0 18 December 1998 (18.12.98) EP

(71) Applicant *(for all designated States except US)*: SMITHKLINE BEECHAM PLC [GB/GB]; New Horizons Court, Brentford, Middlesex TW8 9EP (GB).

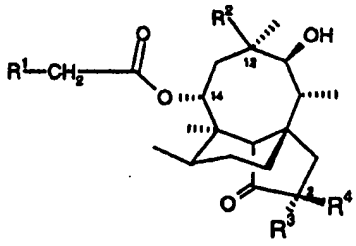
(72) Inventors; and
(75) Inventors/Applicants *(for US only)*: BROOKS, Gerald [GB/GB]; SmithKline Beecham Pharmaceuticals, New Frontiers Science Park South, Third Avenue, Essex CM19 5AW (GB). HUNT, Eric [GB/GB]; SmithKline Beecham Pharmaceuticals, New Frontiers Science Park South, Third Avenue, Essex CM19 5AW (GB).

(74) Agent: CONNELL, Anthony, Christopher; Corporate Intellectual Property, SmithKline Beecham, Two New Horizons Court, Brentford, Middlesex TW8 9EP (GB).

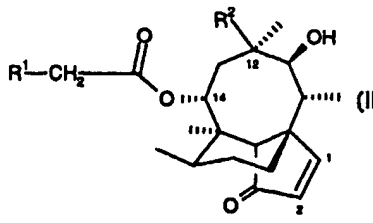
(81) Designated States: AE, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, CA, CH, CN, CR, CU, CZ, DE, DK, DM, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, NO, NZ, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TR, TT, TZ, UA, UG, US, UZ, VN, YU, ZA, ZW, ARIPO patent (GH, GM, KE, LS, MW, SD, SL, SZ, TZ, UG, ZW), Eurasian patent (AM, AZ, BY, KG, KZ, MD, RU, TJ, TM), European patent (AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE), OAPI patent (BF, BJ, CF, CG, CI, CM, GA, GN, GW, ML, MR, NE, SN, TD, TG).

Published
With international search report.
Before the expiration of the time limit for amending the claims and to be republished in the event of the receipt of amendments.

(54) Title: MUTILIN 14-ESTER DERIVATIVES HAVING ANTIBACTERIAL ACTIVITY



(IA)



(IB)

(57) Abstract

A compound of formula (IA) or (IB) in which R^1 is an optionally substituted heteroaryl group which comprises a five membered heteroaromatic ring which has at least one nitrogen atom and which is linked via a nitrogen atom; R^2 is vinyl or ethyl; R^3 is H, OH or F, and R^4 is H, or R^3 is H and R^4.

FOR THE PURPOSES OF INFORMATION ONLY

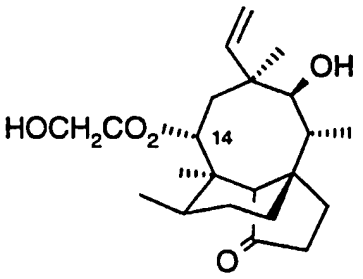
Codes used to identify States party to the PCT on the front pages of pamphlets publishing international applications under the PCT.

AL	Albania	**ES**	Spain	**LS**	Lesotho	**SI**	Slovenia
AM	Armenia	**FI**	Finland	**LT**	Lithuania	**SK**	Slovakia
AT	Austria	**FR**	France	**LU**	Luxembourg	**SN**	Senegal
AU	Australia	**GA**	Gabon	**LV**	Latvia	**SZ**	Swaziland
AZ	Azerbaijan	**GB**	United Kingdom	**MC**	Monaco	**TD**	Chad
BA	Bosnia and Herzegovina	**GE**	Georgia	**MD**	Republic of Moldova	**TG**	Togo
BB	Barbados	**GH**	Ghana	**MG**	Madagascar	**TJ**	Tajikistan
BE	Belgium	**GN**	Guinea	**MK**	The former Yugoslav Republic of Macedonia	**TM**	Turkmenistan
BF	Burkina Faso	**GR**	Greece			**TR**	Turkey
BG	Bulgaria	**HU**	Hungary	**ML**	Mali	**TT**	Trinidad and Tobago
BJ	Benin	**IE**	Ireland	**MN**	Mongolia	**UA**	Ukraine
BR	Brazil	**IL**	Israel	**MR**	Mauritania	**UG**	Uganda
BY	Belarus	**IS**	Iceland	**MW**	Malawi	**US**	United States of America
CA	Canada	**IT**	Italy	**MX**	Mexico	**UZ**	Uzbekistan
CF	Central African Republic	**JP**	Japan	**NE**	Niger	**VN**	Viet Nam
CG	Congo	**KE**	Kenya	**NL**	Netherlands	**YU**	Yugoslavia
CH	Switzerland	**KG**	Kyrgyzstan	**NO**	Norway	**ZW**	Zimbabwe
CI	Côte d'Ivoire	**KP**	Democratic People's Republic of Korea	**NZ**	New Zealand		
CM	Cameroon			**PL**	Poland		
CN	China	**KR**	Republic of Korea	**PT**	Portugal		
CU	Cuba	**KZ**	Kazakstan	**RO**	Romania		
CZ	Czech Republic	**LC**	Saint Lucia	**RU**	Russian Federation		
DE	Germany	**LI**	Liechtenstein	**SD**	Sudan		
DK	Denmark	**LK**	Sri Lanka	**SE**	Sweden		
EE	Estonia	**LR**	Liberia	**SG**	Singapore		

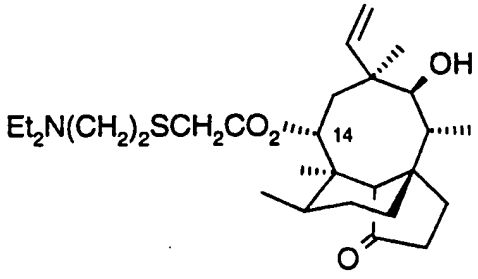
MUTILIN 14-ESTER DERIVATIVES HAVING ANTIBACTERIAL ACTIVITY

The present invention relates to novel compounds, to processes for their preparation, to pharmaceutical compositions containing them and to their use in medical therapy, particularly antibacterial therapy.

Pleuromutilin, the compound of formula (A), is a naturally occurring antibiotic which has antimycoplasmal activity and modest antibacterial activity. Mutilin and other compounds with a free OH at C-14 are inactive. The impact of further modification at C-14 on the activity of pleuromutilin has been investigated (H. Egger and H. Reinshagen, *J Antibiotics*, 1976, 29, 923). Replacing the hydroxy group of the glycolic ester moiety at position 14 by another O, S or N-linked group was found to improve anti-microbial activity. Thus, introducing a diethylaminoethylthio group gives the compound of formula (B), also known as Tiamulin, which is used as a veterinary antibiotic (G. Hogenauer in *Antibiotics*, Vol. V, part 1, ed. F.E. Hahn, Springer-Verlag, 1979, p.344). Further examples of such compounds included *inter alia* two N-linked non-aromatic heterocyclyl acetates, a piperazinoacetate and a morpholinoacetate.



(A)



(B)

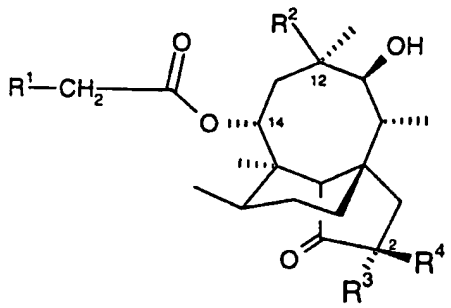
In this application, the non-conventional numbering system which is generally used in the literature (G. Hogenauer, *loc.cit.*) is used.

WO 97/25309 (SmithKline Beecham) describes further modification of the acyloxy group, disclosing 14-*O*-carbamoyl derivatives of mutilin or 19, 20-dihydromutilin, in which the N-atom of the carbamoyl group is unsubstituted, mono- or di-substituted.

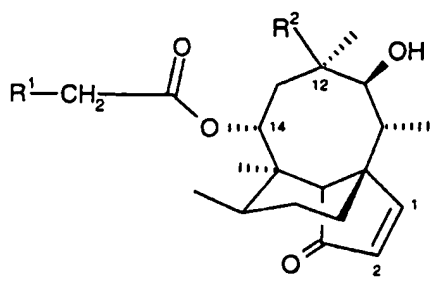
WO 98/05659 (SmithKline Beecham) discloses 14-*O*-carbamoyl derivatives of mutilin or 19, 20-dihydromutilin, in which the N-atom of the carbamoyl group is acylated by a group which includes an azabicyclic moiety. WO 99/21855 (SmithKline Beecham) discloses mono- and bicyclic N-containing ester moieties on the C-14 position of mutilin or 19, 20-dihydromutilin. International Application No. PCT/GB/99/02575 (SmithKline Beecham) discloses 14-acyloxy derivative of mutilin or 19,20-dihydromutilin having a 2-fluoro substituent.

The present invention is based on the unexpected discovery that novel mutilin derivatives having a heteroaryl acetate substituent at the 14-position also have potent antimicrobial activity.

Accordingly the present invention provides a compound of formula (IA) or (IB):



(IA)



(IB)

in which:

R^1 is an optionally substituted heteroaryl group which comprises a five membered heteroaromatic ring which has at least one nitrogen atom and which is linked via a nitrogen atom;

R^2 is vinyl or ethyl; and

R^3 is H, OH or F, and R^4 is H, or R^3 is H and R^4 is F.

In the heteroaryl group R^1, the five membered ring may be fused to a second ring having from 4 to 7, preferably 5 or 6, ring atoms, which ring may comprise up to four, preferably 1 or 2, heteroatoms each selected from oxygen, nitrogen and sulphur and which may be a (hetero)aromatic or a non-aromatic carbocyclic or heterocyclic ring.

Representative values for the heteroaryl group of R^1 include pyrrole, pyrazole, imidazole, 1,2,3-triazole, 1,2,4-triazole, tetrazole, indole, benzimidazole, benzotriazole, 2-aza-indole and 6-aza-indole. Preferably, the heteroaryl group is pyrrole or pyrazole.

Suitably, R^1 may include up to three substitutents. Representative substituents include (C_{1-6})alkyl, hydroxy(C_{1-6})alkyl, amino(C_{1-6})alkyl, mono- and di-*N*-(C_{1-6})alkylamino(C_{1-6})alkyl, the residue of an α-amino acid, acyloxy, formyl, carboxy and salt, ester and amide derivatives thereof, carboxy(C_{1-6})alkyl and salt and ester derivatives thereof, mono- and di-*N*-(C_{1-6})alkylcarbamoyl, arylcarbamoyl, amidino, aryl, aryl(C_{1-4})alkyl, heterocyclyl, and heterocyclyl(C_{1-6})alkyl. Suitable ester and amide derivatives of a carboxy group include those comprising (C_{1-6})alkyl, aryl or heterocyclyl groups. Suitable heterocyclo/yl rings for use as substituents include those having a basic ring, for instance those comprising a nitrogen atom such as piperidinyl, morpholinyl, and pyrrolidinyl which ring may be optionally substituted.

Preferred substituents include a basic amine functionality, for instance mono- or di-(C_{1-6})alkylamino, for example dimethylamino; mono- or di-(C_{1-6})alkylaminoalkyl, for example 2-aminoethyl, 2-dimethylaminoethyl; N-heterocyclyl, for example piperidinyl; and N-heterocyclo-(C_{1-6})alkyl, for example piperidinomethyl, morpholinomethyl and pyrrolidinomethyl.

Alkyl and alkenyl groups referred to herein (individually or as part of alkoxy or alkenyloxy) may be straight and branched groups containing up to six carbon atoms and are optionally substituted by one or more groups selected from the group consisting of aryl, heteroaryl, heterocyclyl, (C_{1-6})alkoxy, (C_{1-6})alkylthio, aryl(C_{1-6})alkoxy, aryl(C_{1-6})alkylthio, amino, mono- or di-(C_{1-6})alkylamino, cycloalkyl, cycloalkenyl, carboxy and esters thereof, amide, ureido, guanidino, (C_{1-6})alkylguanidino, amidino, (C_{1-6})alkylamidino, (C_{1-6})acyloxy, azido, hydroxy, and halogen.

Cycloalkyl and cycloalkenyl groups referred to herein include groups having from three to eight ring carbon atoms and are optionally substituted as described hereinabove for alkyl and alkenyl groups.

When used herein, the term "aryl" means single and fused rings suitably containing from 4 to 7, preferably 5 or 6, ring atoms in each ring, which rings, may each be unsubstituted or substituted by, for example, up to three substituents. A fused ring system may include aliphatic rings and need include only one aromatic ring. Suitable aryl groups include phenyl and naphthyl such as 1-naphthyl or 2-naphthyl.

Suitably any aryl group, including phenyl and naphthyl, may be optionally substituted by up to three, preferably up to three substituents. Suitable substituents include halogen, (C_{1-6})alkyl, aryl, aryl(C_{1-6})alkyl, (C_{1-6})alkoxy, (C_{1-6})alkoxy(C_{1-6})alkyl, halo(C_{1-6})alkyl, aryl(C_{1-6})alkoxy, hydroxy, nitro, cyano, azido, amino, mono- and di-*N*-(C_{1-6})alkylamino, acylamino, arylcarbonylamino, acyloxy, carboxy, carboxy salts, carboxy esters, carbamoyl, mono- and di-*N*-(C_{1-6})alkylcarbamoyl, (C_{1-6})alkoxycarbonyl, aryloxycarbonyl, ureido, guanidino, (C_{1-6})alkylguanidino, amidino, (C_{1-6})alkylamidino, sulphonylamino, aminosulphonyl, (C_{1-6})alkylthio, (C_{1-6})alkyl sulphinyl, (C_{1-6})alkylsulphonyl, heterocyclyl, heteroaryl, heterocyclyl(C_{1-6})alkyl and heteroaryl(C_{1-6})alkyl. In addition, two adjacent ring carbon atoms may be linked by a (C_{3-5})alkylene chain, to form a carbocyclic ring.

When used herein the terms "heterocyclyl" and "heterocyclic" suitably include, unless otherwise defined, non-aromatic, single and fused, rings suitably containing up to four heteroatoms in each ring, each of which is selected from oxygen, nitrogen and sulphur, which rings, may be unsubstituted or substituted by, for example, up to three substituents. Each heterocyclic ring suitably has from 4 to 7, preferably 5 or 6, ring

atoms. A fused heterocyclic ring system may include carbocyclic rings and need include only one heterocyclic ring.

When used herein, the term "heteroaryl" suitably include, unless otherwise defined, a mono- or bicyclic heteroaromatic ring system comprising up to four, preferably 1 or 2, heteroatoms each selected from oxygen, nitrogen and sulphur. Each ring may have from 4 to 7, preferably 5 or 6, ring atoms. A bicyclic heteroaromatic ring system may include a carbocyclic ring. When substituted, a heteroaryl group may comprise up to three substituents.

Preferably a substituent for a heterocyclyl or a heteroaryl group is selected from halogen, (C_{1-6})alkyl, aryl(C_{1-6})alkyl, (C_{1-6})alkoxy, (C_{1-6})alkoxy(C_{1-6})alkyl, halo(C_{1-6})alkyl, hydroxy, amino, mono- and di-*N*-(C_{1-6})alkyl-amino, acylamino, carboxy, carboxy salts, carboxy esters, carbamoyl, mono- and di-*N*-(C_{1-6})alkylcarbonyl, aryloxycarbonyl, (C_{1-6})alkoxycarbonyl(C_{1-6})alkyl, aryl, oxy groups, ureido, guanidino, (C_{1-6})alkylguanidino, amidino, (C_{1-6})alkylamidino, sulphonylamino, aminosulphonyl, (C_{1-6})alkylthio, (C_{1-6})alkylsulphinyl, (C_{1-6})alkylsulphonyl, heterocyclyl, heteroaryl, heterocyclyl(C_{1-6})alkyl and heteroaryl(C_{1-6})alkyl.

Depending on the substituents, two or more diastereoisomers may be possible. In that situation the present invention includes the individual diastereoisomers and mixtures thereof.

The 2-hydroxy-substituted compounds of formula (IA) are of the (2*S*) configuration. The 2-F-substituted compounds of formula (IA) may of (2*S*) configuration or (2*R*) configuration, or be provided as mixtures thereof. The (2*S*) configuration is however preferred.

Preferred compounds of the invention include:

[3-(Pyrrolidin-1-ylmethyl)pyrrol-1-yl]acetic acid mutilin 14-ester;

[3-(Carboxamidomethyl)indol-1-yl]acetic acid mutilin 14-ester;

[3-(2-Aminoethyl)indol-1-yl]acetic acid mutilin 14-ester;

[4-(1-Methyl-1,2,3,6-tetrahydropyridin-4-yl)pyrazol-1-yl]acetic acid mutilin 14-ester;

[5-(Piperidin-4-yl)pyrazol-1-yl]acetic acid mutilin 14-ester;

[3-(Piperidin-4-yl)pyrazol-1-yl]acetic acid mutilin 14-ester;

[4-(Piperidin-4-yl)pyrazol-1-yl]acetic acid 19,20-dihydro-mutilin 14-ester;

{3-[1-(1,2-Dioxo–3-dimethylamino-3-cyclobuten-4-yl)piperidin-4-yl]pyrazol-1-yl}acetic acid mutilin 14-ester; and

(Pyrazol-1-yl)acetic acid mutilin 14-ester.

The compounds of this invention may be in crystalline or non-crystalline form, and, if crystalline, may optionally be hydrated or solvated. This invention includes within

its scope stoichiometric hydrates as well as compounds containing variable amounts of water.

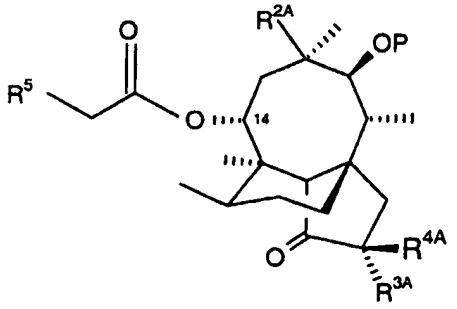
The compounds according to the invention are suitably provided in substantially pure form, for example at least 50% pure, suitable at least 60% pure, advantageously at least 75% pure, preferably at least 85% pure, more preferably at least 95% pure, especially at least 98% pure, all percentages being calculated as weight/weight.

Compounds of the invention that contain a basic group such as an amino substituent, may be in the form of a free base or an acid addition salt. Compounds having an acidic group such as a carboxy substituent may be in the form of a pharmaceutically acceptable salt. Compounds of the invention having both a basic and an acidic centre may be in the form of zwitterions, acid addition salt of the basic centre or alkali metal salts (of the carboxy group). Pharmaceutically acceptable salts are preferred.

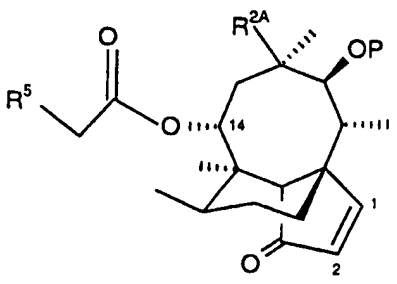
Pharmaceutically acceptable acid-addition salts include those described by Berge, Bighley, and Monkhouse, *J. Pharm. Sci.*, 1977, 66, 1-19. Suitable salts include the hydrochloride, maleate, and methanesulphonate; particularly the hydrochloride.

Pharmaceutically acceptable salts include those described by Berge, Bighley, and Monkhouse, *J. Pharm. Sci.*, 1977, 66, 1-19. Suitable salts include alkali metal salts such as the sodium and potassium salts.

Compounds of the present invention may be readily prepared from a pleuromutilin or a 19,20-dihydro-pleuromutilin derivative by adapting procedures well known in the art for alkylating the nitrogen of a 5-membered heteroaryl group. Accordingly, the present invention provides a process for preparing a compound of formula (IA) or (IB) which comprises reacting a compound of formula (IIA) or (IIB):



(IIA)



(IIB)

in which:

R^5 is a leaving group or hydroxy, P is hydrogen or a removable hydroxy-protecting group, and R^{2A}, R^{3A} and R^{4A} are R^2, R^3 and R^4 as defined for formulae (IA) and (IB) or groups convertible to R^2, R^3 and R^4;

(i) when R^5 is a leaving group, with R^1H under alkylating conditions; or
(ii) when R^5 is a hydroxy, with $R^{1A}H$ in which R^{1A} is a group R^1 which contains an acidic NH, under Mitsunobu conditions;
and thereafter, and if so needed;
converting P to hydrogen,
converting an R^{2A}, R^{3A} or R^{4A} group to an R^2, R^3 or R^4 group, and/or
converting one R^1, R^2, R^3or R^4 group to another R^1, R^2, R^3or R^4 group.

Suitable values of R^5 include mesylate, tosylate, trifluoromenthane sulphonate, chloride, bromide, and iodide.

Suitable alkylating conditions include an organic solvent such as *N,N*-dimethylformamide, dimethylsulfoxide, tetrahydrofuran, or acetonitrile (preferably *N,N*-dimethylformamide), at a temperature of –20°C to 60°C (preferably 0°C to 23°C), and in the presence of an inorganic base such as sodium hydride, potassium hydride, lithium hydride, or potassium carbonate (preferably sodium hydride).

When the 5-membered nitrogen heteroaryl ring of R^1 contains a sufficiently acidic NH group (*e.g.* pKa < 10), for example tetrazole, a compound of formula (IA) or (IB) may be produced using a Mitsunobu reaction (O. Mitsunobu, *Synthesis*, 1981, 1). For example, $R^{1A}H$ (*e.g.* tetrazole) is brought into reaction with pleuromutilin or 19,20-dihydro-pleuromutilin in the presence of an azodicarboxylate (*e.g.* diethyl azodicarboxylate) and a triaryl or trialkyl phosphine (*e.g.* triphenyl phosphine) in a solvent such as tetrahydrofuran or acetonitrile at a temperature of 0°C to 25°C.

It will be appreciated that the above methods may provide a mixture of positional isomers. If, for instance, R^1H is a 4-substituted imidazole, this may give a mixture of 4- and 5-substituted *N*-linked imidazole acetates. Such mixtures may be resolved by conventional separation techniques such as chromatography, fractional crystallisation etc.

Conversions of an R^{2A}, R^{3A}or R^{4A} group to an R^2, R^3or R^4 group typically arise when a protecting group is needed during the above coupling reaction or during the preparation of the reactants by the procedures described below. Interconversion of one R^1, R^2, R^3 or R^4 group to another typically arises when one compound of formula (IA/B) is used as the immediate precursor of another compound of formula (IA/B) or when it is easier to introduce a more complex or reactive substituent at the end of a synthetic sequence. A substituent group in R^1 can be converted into another substituent group using one of the general methods for functional group transformation described in the literature (*e.g.* a carboxylic ester can be hydrolysed to a carboxylic acid with base; an acid can be converted into an amide; a *tert*-butoxy-carbonyl-amino group can be converted into an amine by treatment with trifluoroacetic acid; an amino group can be

alkylated or acylated), provided that the method chosen is compatible with other functional groups in the molecule (*e.g.* the ketone at C-3 in the pleuromutilin nucleus).

Functional group transformations are well known in the art and are described in, for instance, Comprehensive Organic Functional Group Transformations, eds. A.R. Katritzky, O. Meth-Cohn, and C.W. Rees (Elsevier Science Ltd., Oxford, 1995), Comprehensive Organic Chemistry, eds. D. Barton and W.D. Ollis (Pergamon Press, Oxford, 1979), and Comprehensive Organic Transformations, R.C. Larock (VCH Publishers Inc., New York, 1989).

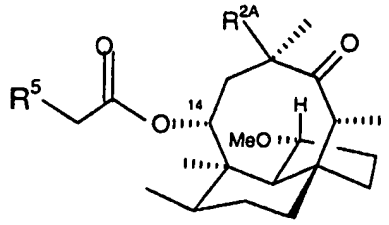
Preferably P is a hydroxyl protecting group such as an acyl group, for example so that -OP is trifluoroacetoxy or dichloroacetoxy. When the intended R^3 is also hydroxyl, then R^{3A} is also preferably acyloxy, for example acetyl or dichloroacetyl. Hydroxyl groups at positions 11 and 2 (as groups OP and R^{3A}) may be protected using, for example, dichloroacetic anhydride and pyridine in tetrahydrofuran or *N*-trifluoroacetyl-imidazole in tetrahydrofuran at 0°C. After the reaction described above with $R^1H/R^{1A}H$ is complete, the protecting acyl groups may be removed to restore the hydroxyl groups, for instance by hydrolysis *e.g.* using NaOH in MeOH.

Suitable hydroxy, carboxy and amino protecting groups are those well known in the art and which may be removed under conventional conditions and without disrupting the remainder of the molecule. A comprehensive discussion of the ways in which hydroxy, carboxy and amino groups may be protected and methods for cleaving the resulting protected derivatives is given in for example "Protective Groups in Organic Chemistry" (T.W. Greene and P.G.M. Wuts, Wiley-Interscience, New York, 2nd edition, 1991). Particularly suitable hydroxy protecting groups include, for example, triorganosilyl groups such as, for instance, trialkylsilyl and also organocarbonyl and organooxycarbonyl groups such as, for instance, acetyl, allyloxycarbonyl, 4-methoxybenzyloxycarbonyl and 4-nitrobenzyloxycarbonyl. Particularly suitable carboxy protecting groups include alkyl and aryl groups, for instance methyl, ethyl and phenyl. Particularly suitable amino protecting groups include alkoxycarbonyl, 4-methoxybenzyloxycarbonyl and 4-nitrobenzyloxycarbonyl.

R^{2A} is typically the R^2 group vinyl, and this may be converted to the alternative R^2 ethyl group by hydrogenating the vinyl group to form an ethyl group, typically by hydrogenation over a palladium catalyst (*e.g.* 10% palladium-on-carbon) in a solvent such as ethyl acetate, ethanol, dioxane, or tetrahydrofuran.

R^{3A} is typically hydrogen, fluoro or protected hydroxyl, such as acyloxy. After the coupling reaction, protecting acyl groups may be removed to restore the hydroxyl groups by hydrolysis *e.g.* using NaOH in MeOH.

A compound of formula (IA) may also be prepared from an *epi*-mutilin starting material. Accordingly, in a further aspect, the present invention provides a process for preparing a compound of formula (IA) in which R^3 and R^4 are both hydrogen which comprises reacting an *epi*-mutilin compound of formula (IIC):



(IIC)

where:

R^{2A} and R^5 are as defined for formulae (IIA) and (IIB);

with a compound R^1H or $R^{1A}H$, as hereinbefore described, and then

treating the product with an acid,

and where required or desired

converting an R^{2A} group to a R^2 group, and/or

converting one R^1 or R^2 group to another R^1 or R^2 group.

The acid treatment indicated above converts the *epi*-mutilin configuration of formula (IIC) to the usual mutilin nucleus of formula (IIA). Typically this conversion is carried out by treatment with conc. HCl or Lukas reagent (conc. HCl saturated with $ZnCl_2$) in dioxane.

As in formulae (IIA) and (IIB), R^{2A} is typically the R^2 group vinyl, and this may be converted to the alternative R^2 group by hydrogenating the vinyl group to form an ethyl group.

Compounds of formulae (IIA) in which R^{3A} and R^{4A} are hydrogen, (IIB) and (IIC) may be readily prepared from pleuromutilin (R^{2A} = vinyl, R^5 = OH) or 19,20-dihydro-pleuromutilin (R^2 = ethyl, R^3 = OH) by methods described in the literature and in WO 97/25309 and WO 98/05659 (SmithKline Beecham).

Compounds of formula (IIA) in which R^{3A} is hydroxyl or fluoro may be readily prepared from pleuromutilin, via an intermediate 2-diazo compound, the preparation of which is described by G. Schulz and H. Berner in *Tetrahedron*, 1984, 40, 905.

The intermediate 2-diazo compound may be reacted with a carboxylic acid to give a 2-acyloxy-mutilin derivative, effectively a compound of formula (IIA) in which R^{3A} is protected hydroxyl. Suitably, reaction with dichloroacetic acid gives 2-dichloroacetoxy-mutilin derivative, which can be deprotected as described above to provide the (2*S*)-2-hydroxy derivative, at an appropriate stage.

Compounds of formula (IIA) in which R^{3A} is fluoro may be obtained by reacting 2-diazo-mutilin with a source of hydrogen fluoride. Conveniently, the hydrogen fluoride source is an amine complex of hydrogen fluoride such as hydrogen fluoride-pyridine. The reaction may be carried out in an anhydrous solvent (*e.g.* diethyl ether, tetrahydrofuran, 1,2-dimethoxyethane), at a temperature of -15°C to 25°C. This reaction produces (2*S*)-2-fluoro derivatives. (2*R*)-2-Fluoro-mutilin derivatives may be prepared by treating the (2*S*)-isomer with a base (*e.g.* sodium hydroxide or potassium hydroxide in ethanol). This will usually produce a mixture of (2*S*)- and (2*R*)-isomers that may be separated using conventional techniques such as chromatography and crystallisation.

The compounds of the present invention may contain a chiral centre, and therefore the above processes may produce a mixture of diastereoisomers. A single diastereoisomer may be prepared by separating such a mixture of diastereoisomers which has been synthesised using a racemic starting material, or by synthesis using an optically pure starting material.

The compounds of this invention may be in crystalline or non-crystalline form, and, if crystalline, may optionally be hydrated or solvated. When some of the compounds of this invention are allowed to crystallise or are recrystallised from organic solvents, solvent of crystallisation may be present in the crystalline product. Similarly, some of the compounds of this invention may be crystallised or recrystallised from solvents containing water. In such cases water of hydration may be present in the crystalline product. Crystallisation procedures will usually produce stoichiometric hydrates. Compounds containing variable amounts of water may be produced by processes such as lyophilisation.

The compounds according to the invention are suitably provided in substantially pure form, for example at least 50% pure, suitable at least 60% pure, advantageously at least 75% pure, preferably at least 85% pure, more preferably at least 95% pure, especially at least 98% pure, all percentages being calculated as weight/weight. An impure or less pure form of a compound according to the invention may, for example, be used in the preparation of a more pure form of the same compound or of a related compound (for example a corresponding derivative) suitable for pharmaceutical use.

The present invention also includes pharmaceutically acceptable salts and derivatives of the compounds of the invention. Salt formation may be possible when one of the substituents carries an acidic or basic group. Salts may be prepared by salt exchange in conventional manner.

Acid-addition salts may be pharmaceutically acceptable or non-pharmaceutically acceptable. In the latter case, such salts may be useful for isolation and purification of the

compound of the invention, or intermediates thereto, and will subsequently be converted into a pharmaceutically acceptable salt or the free base.

The compounds of the present invention and their pharmaceutically acceptable salts or derivatives have antimicrobial properties and are therefore of use in therapy, in particular for treating microbial infections in animals, especially mammals, including humans, in particular humans and domesticated animals (including farm animals). The compounds may be used for the treatment of infections caused by, for example, Gram-positive and Gram-negative bacteria and mycoplasmas, including, for example, *Staphylococcus aureus*, *Staphylococcus epidermidis*, *Enterococcus faecalis*, *Streptococcus pyogenes*, *Streptococcus agalactiae*, *Streptococcus pneumoniae*, *Haemophilus sp.*, *Neisseria sp.*, *Legionella spp.*, *Chlamydia spp.*, *Moraxella catarrhalis*, *Mycoplasma pneumoniae*, and *Mycoplasma gallisepticum*.

The present invention also provides a method of treating microbial infections in animals, especially in humans and in domesticated mammals, which comprises administering a compound of the invention or a pharmaceutically acceptable salt or derivative or solvate thereof, or a composition according to the invention, to a patient in need thereof.

The invention further provides the use of a compound of the invention or a pharmaceutically acceptable salt or derivative or solvate thereof in the preparation of a medicament for use in the treatment of microbial infections.

Compounds of the present invention may be used to treat skin and soft tissue infections and acne, by topical application. Accordingly, in a further aspect the present invention provides the use of a compound of the invention or a pharmaceutically acceptable salt or derivative or solvate thereof in the preparation of a medicament adapted for topical administration for use in the treatment of skin and soft tissue infections and also in the treatment of acne in humans.

Compounds of the present invention may be also used for the elimination or reduction of nasal carriage of pathogenic bacteria such as *S. aureus*, *H. influenzae*, *S. pneumonia* and *M. catarrhalis*, in particular colonisation of the nasospharynx by such organisms, by the administration of a compound of the present invention thereto. Accordingly, in a further aspect, the present invention provides for the use of a compound of the invention or a pharmaceutically acceptable salt or derivative or solvate thereof in the manufacture of a medicament adapted for administration to the nasal cavity, for reducing or eliminating the nasal carriage of pathogenic organisms. Preferably, the medicament is adapted for focussed delivery to the nasopharynx, in particular the anterior nasopharynx.

Such reduction or elimination of nasal carriage is believed to be useful in prophylaxis of recurrent acute bacterial sinusitis or recurrent otitis media in humans, in particular in reducing the number of episodes experienced by a patient over a given period of time or increasing the time intervals between episodes. Accordingly, in a further aspect, the present invention provides for the use of a compound of the invention or a pharmaceutically acceptable salt or derivative or solvate thereof in the manufacture of a medicament adapted for administration to the nasal cavity, for prophylaxis of recurrent acute bacterial sinusitis or recurrent otitis media.

Compounds of the present invention are also useful in treating chronic sinusitis. Accordingly, in a further aspect, the present invention provides for the use of a compound of the invention or a pharmaceutically acceptable salt or derivative or solvate thereof in the manufacture of a medicament, for treating of chronic sinusitis.

The compounds according to the invention may suitably be administered to the patient at a daily dosage of from 1.0 to 50 mg/kg of body weight. For an adult human (of approximately 70 kg body weight), from 50 to 3000 mg, for example about 1500 mg, of a compound according to the invention may be administered daily. Suitably, the dosage for adult humans is from 5 to 20 mg/kg per day. Higher or lower dosages may, however, be used in accordance with normal clinical practice.

To lessen the risk of encouraging the development of resistant organisms during prophylaxis of recurrent otitis media or recurrent acute bacterial sinusitis, it is preferred to administer the drug on an intermittent, rather than a continual, basis. In a suitable intermittent treatment regimen for prophylaxis of recurrent otitis media or recurrent sinusitis, drug substance is administered on a daily basis, for a small number of days, for instance from 2 to 10, suitably 3 to 8, more suitably about 5 days, the administration then being repeated after an interval, for instance, on a monthly basis over a period of months, for instance up to six months. Less preferably, the drug substance may be administered on a continuing, daily basis, over a prolonged period, for instance several months. Suitably, for prophylaxis of recurrent otitis media or recurrent sinusitis, drug substance is administered once or twice a day. Suitably, drug substance is administered during the winter months when bacterial infections such as recurrent otitis media and recurrent sinusitis tend to be more prevalent. The drug substance may be administered at a dosage of from 0.05 to 1.00mg, typically about 0.1 to 0.2mg, in each nostril, once or twice a day.

More generally, the compounds and compositions according to the invention may be formulated for administration in any convenient way for use in human or veterinary medicine, by analogy with other antibiotics.

Accordingly, in a further aspect, the present invention provides a pharmaceutical composition comprising a compound of the invention or a pharmaceutically acceptable salt or derivative or solvate thereof together with a pharmaceutically acceptable carrier or excipient.

The compounds and compositions according to the invention may be formulated for administration by any route, for example oral, topical or parenteral. The compositions may, for example, be made up in the form of tablets, capsules, powders, granules, lozenges, creams, syrups, sprays or liquid preparations, for example solutions or suspensions, which may be formulated for oral use or in sterile form for parenteral administration by injection or infusion.

Tablets and capsules for oral administration may be in unit dosage form, and may contain conventional excipients including, for example, binding agents, for example, syrup, acacia, gelatin, sorbitol, tragacanth, or polyvinylpyrrolidone; fillers, for example lactose, sugar, maize-starch, calcium phosphate, sorbitol or glycine; tabletting lubricants, for example magnesium stearate, talc, polyethylene glycol or silica; disintegrants, for example potato starch; and pharmaceutically acceptable wetting agents, for example sodium lauryl sulphate. The tablets may be coated according to methods well known in normal pharmaceutical practice.

Oral liquid preparations may be in the form of, for example, aqueous or oily suspensions, solutions, emulsions, syrups or elixirs, or may be presented as a dry product for reconstitution with water or another suitable vehicle before use. Such liquid preparations may contain conventional additives, including, for example, suspending agents, for example sorbitol, methyl cellulose, glucose syrup, gelatin, hydroxyethyl cellulose, carboxymethyl cellulose, aluminium stearate gel or hydrogenated edible fats; emulsifying agents, for example lecithin, sorbitan monooleate or acacia; non-aqueous vehicles (which may include edible oils), for example almond oil, oily esters (for example glycerine), propylene glycol, or ethyl alcohol; preservatives, for example methyl or propyl p-hydroxybenzoate or sorbic acid; and, if desired, conventional flavouring and colour agents.

Compositions according to the invention intended for topical administration may, for example, be in the form of ointments, creams, lotions, eye ointments, eye drops, ear drops, nose drops, nasal sprays, impregnated dressings, and aerosols, and may contain appropriate conventional additives, including, for example, preservatives, solvents to assist drug penetration, and emollients in ointments and creams. Such topical formulations may also contain compatible conventional carriers, for example cream or ointment bases, ethanol or oleyl alcohol for lotions and aqueous bases for sprays. Such

carriers may constitute from about 1% to about 98% by weight of the formulation: more usually they will constitute up to about 80% by weight of the formulation.

Compositions according to the invention intended for topical administration, in addition to the above, may also contain a steroidal anti-inflammatory agent; for example, betamethasone.

Compositions according to the invention may be formulated as suppositories, which may contain conventional suppository bases, for example cocoa-butter or other glycerides.

Compositions according to the invention intended for parenteral administration may conveniently be in fluid unit dosage forms, which may be prepared utilizing the compound and a sterile vehicle, water being preferred. The compound, depending on the vehicle and concentration used, may be either suspended or dissolved in the vehicle. In preparing solutions, the compound may be dissolved in water for injection and filter-sterilised before being filled into a suitable vial or ampoule, which is then sealed. Advantageously, conventional additives including, for example, local anaesthetics, preservatives, and buffering agents can be dissolved in the vehicle. In order to enhance the stability of the solution, the composition may be frozen after being filled into the vial, and the water removed under vacuum; the resulting dry lyophilised powder may then be sealed in the vial and a accompanying vial of water for injection may be supplied to reconstitute the liquid prior to use. Parenteral suspensions may be prepared in substantially the same manner except that the compound is suspended in the vehicle instead of being dissolved and sterilisation cannot be accomplished by filtration. The compound may instead be sterilised by exposure to ethylene oxide before being suspended in the sterile vehicle. Advantageously, a surfactant or wetting agent is included in such suspensions in order to facilitate uniform distribution of the compound.

A compound or composition according to the invention is suitably administered to the patient in an antimicrobially effective amount.

A composition according to the invention may suitably contain from 0.001% by weight, preferably (for other than spray compositions) from 10 to 60% by weight, of a compound according to the invention (based on the total weight of the composition), depending on the method of administration.

When the compositions according to the invention are presented in unit dosage form, for instance as a tablet, each unit dose may suitably comprise from 25 to 1000 mg, preferable from 50 to 500 mg, of a compound according to the invention.

Representative compositions of the present invention include those adapted for intranasal administration, in particular, those that will reach into the nasopharynx. Such

compositions are preferably adapted for focussed delivery to, and residence within, the nasopharynx. The term "focussed delivery" is used to mean that the composition is delivered to the nasopharynx, rather than remaining within the nares. The term "residence" within the nasopharynx is used to mean that the composition, once delivered to the nasopharynx, remains within the nasopharynx over a course of several hours, rather than being washed away more or less immediately. Preferred compositions include spray compositions and creams. Representative spray compositions include aqueous compositions, as well as oily compositions which contain amphiphilic agents so that the composition increases in viscosity when in contact with moisture. Creams may also be used, especially creams having a rheology that allows the cream to spread readily in the nasopharynx.

Preferred aqueous spray compositions include, in addition to water, further excipients including a tonicity modifier such as a salt, for instance sodium chloride; preservative, such as benzalkonium salt; a surfactant such as a non-ionic surfactant, for instance a polysorbate; and buffer, such as sodium dihydrogen phosphate; present in low levels, typically less than 1%. The pH of the composition may also be adjusted, for optimum stability of the drug substance during storage. For compounds of the present invention, a pH in the range 5 to 6, preferably about 5.3 to 5.8, typically about 5.5 is optimal.

Representative oily spray and cream compositions are described in WO 98/14189 (SmithKline Beecham). Representative aqueous sprays are described in WO 99/21855 (SmithKline Beecham).

Suitably, the drug substance is present in compositions for nasal delivery in between 0.001 and 5%, preferably 0.005 and 3%, by weight of the composition. Suitable amounts include 0.5% and 1% by weight of the composition (for oily compositions and creams) and from 0.01 to 0.2% (aqueous compositions).

Spray compositions according to the present invention may be delivered to the nasal cavity by spray devices well known in the art for nasal sprays, for instance an air lift pump. Preferred devices include those which are metered to provide a unit volume of composition, preferably about 100µl, and optionally adapted for nasal administration by addition of a modified nozzle.

The invention is illustrated by the following Examples.

Preparation 1 - 4-Hydroxymethylpyrazole - To a solution of ethyl 4-pyrazolecarboxylate (0.73g) in THF (45ml) was added lithium aluminium hydride (0.59g) and the mixture refluxed for 6 hours, cooled and treated with water (2ml) and 2M NaOH solution (4.2ml). After 5 mins. vigorous stirring, the mixture was filtered and the filtrate evaporated to give the title compound (0.3g). In like manner was prepared **3-hydroxy-methyl-5-methylpyrazole.**

Preparation 2 - 3-(2-Aminoethyl)-5-methylpyrazole -

Step 1: 3-Chloromethyl-5-methylpyrazole hydrochloride - A solution of 3-hydroxymethyl-5-methylpyrazole (3.45g) in THF (20ml) was treated with 1M HCl in ether (40ml) and the resulting solid hydrochloride was filtered off and dissolved in thionyl chloride (15ml). The solution was refluxed 15 mins., cooled, evaporated and the residue triturated under ether to give the title compound as a solid (3.86g).

Step 2: 3-Cyanomethyl-5-methylpyrazole - To a vigorously stirred, ice-cooled, part-solution of potassium cyanide (10.2g) in water (12ml) was added dropwise a solution of 3-chloromethyl-5-methylpyrazole hydrochloride (3.86g) in ethanol (45ml). After 1 hour, the reaction was warmed to room temperature, left 4 hours, filtered and evaporated to low volume. Water (15ml) was added and the solution extracted with chloroform (4 x 20ml). The combined extracts were dried and evaporated to give the title compound (2.5g).

Step 3: 3-(2-Aminoethyl)-5-methylpyrazole - A solution of 3-cyanomethyl-5-methylpyrazole (0.85g) in THF (20ml) was treated portionwise with lithium aluminium hydride (0.4g) and stirred overnight. Water (1.4ml) and 2M NaOH solution (1ml) were added and after 5 minutes the solids were filtered off and the filtrate evaporated. Chromatography (chloroform/methanol/35% NH_3 solution 9:1:0.1) provided the title compound (0.3g). MS (+ve ion C.I.): m/z 126 (MH^+, 100%).

Preparation 3 - 3-(1-t-Butoxycarbonylpiperidin-4-yl)pyrazole

Step 1: 3-(Piperidin-4-yl)pyrazole - A mixture of 2-methylene-3-quinuclidinone hydrochloride hydrate (Aldrich, 5g), glycol (15ml), hydrazine hydrate (2.5ml) and potassium hydroxide (5.5g) was heated to 110°C for 2 hours. The temperature was taken up to 200°C while allowing volatiles to distil off and this temperature was then maintained for 10 hours. The mixture was diluted with water (30ml) and extracted with chloroform (10 X 30ml). The chloroform was dried and evaporated to give the title compound (3g). MS (+ve ion electrospray): m/z 152, (MH^+, 100%).

Step 2: 3-(1-t-Butoxycarbonylpiperidin-4-yl)pyrazole - A solution of 3-(piperidin-4-yl)pyrazole (3g) in chloroform (100ml) was treated with di-t-butyldicarbonate (5.5g), left 1 hour, washed with $NaHCO_3$ solution (100 ml), dried and evaporated. Chromatography (75% EtOAc/hexane) provided the title compound as a gum (4.1g).

Preparation 4 - 3-Benzyloxycarbonylpyrrole - A solution of pyrrole-3-carboxylic acid (0.305g) in DMF (15ml) was treated with NaH (60% dispersion, 0.11g), stirred 30 minutes, treated with benzyl bromide (0.36ml) and stirred overnight. The mixture was diluted with EtOAc(40ml), washed with water (3x30ml), dried and evaporated. Chromatography (20% EtOAc/hexane) provided the title compound (0.218g).

Preparation 5 - 3-Pyrrolidin-1-yl)carbonylpyrrole - An ice-cooled solution of pyrole-3-carboxylic acid (0.5g) in DMF(15ml)/dichloromethane (10ml) was treated with pyridine (0.36ml), pyrrolidine (0.38ml) and dichlohexylcarbodimide (1.39g) and stirred overnight at room temperature. The mixture was filtered and the filter washed with $NaHCO_3$ solution (30ml) and water (2 x 30ml), dried and evaporated. Chromatography (50% EtOAc/hexane) gave the title compound (0.17g). MS (+ve ion electrospray): m/z 165, (MH^+, 100%). In similar manner were prepared: **3-(piperidin-1-yl)carbonylpyrrole, 2-(piperidin-1-yl)carbonylindole** and **2-(pyrrolidin-1-yl)carbonylindole.**

Preparation 6 - 3-(Pyrrolidin-1-yl)methylpyrrole - A solution of 3-(pyrrolidin-1-yl)carbonylpyrrole (0.165g) in THF (10ml) was treated with lithium aluminium hydride (0.114g) and refluxed for 2 hours. The mixture was cooled, treated with water (0.4ml) and 2M NaOH solution (0.8ml), stirred vigorously 5 minutes and filtered. Evaporation of the filtrate provided the title compound (0.141g). MS (+ve ion electrospray): m/z 151, (MH^+, 100%). In similar manner were prepared: **3-(piperidin-1-yl)methylpyrrole, 2-(pyrrolidin-1-yl)methylindole, 2-(piperidin-1-yl)methylindole** and **3-(2-aminoethyl)indole** (from indole 3-acetamide).

Preparation 7 – 4-(1-t-butoxycarbonyl-4-hydroxypiperidin-4-yl)pyrazole - A solution of 4-bromopyrazole (1.4g) in THF (30ml) at -78°C was treated dropwise with n-BuLi (6.2ml of 2.7M solution), keeping the temperature below -70°C. Likewise was added t-BuLi (11.8ml of a 1.7M solution). After 20 minutes a solution of 1-t-butoxycarbonyl-4-piperidone (2g) in THF (20ml) was added dropwise, the solution allowed to reach -10°C and kept at this temperature for 2 hours. It was cooled again to -70°C, treated with excess saturated aqueous ammonium chloride, allowed to warm to room temp. and diluted with water to obtain complete solution. THF was evaporated under reduced pressure and the aqueous extracted with EtOAc (2 x 100ml). The organic was dried and evaporated and the residue chromatographed (70% EtOAc/hexane) to give the title compound (1.88g) MS (-ve ion electrospray) m/z 593 (2M OAc^-,100%), 266(M-H^-, 50%).

Preparation 8 - 4-(1,2,3,6-Tetrahydropyridin-4-yl)pyrazole TFA salt - The title compound was prepared from 4-(1-t-butoxycarbonyl-4-hydroxypiperidin-4-yl)pyrazole by treatment with trifluoroacetic acid (TFA) at 65°C for 5 hours (15.1g).

Preparation 9 - 4-(1-t-Butoxycarbonyl-1,2,3,6-tetrahydropyridin-4-yl)pyrazole - 4-(1,2,3,6-Tetrahydropyridin-4-yl)pyrazole TFA salt (2g) in dichloromethane (50ml) was treated with triethylamine (2.2ml) and di-t-butyldicarbonate (1.16g). After 1½ hours the mixture was concentrated *in vacuo*. The residue was dissolved in ethyl acetate, washed with $NaHCO_3$ solution, dried and concentrated *in vacuo*. Chromatography (50% EtOAc/hexane) gave the title compound (0.7g). MS (+ve ion electrospray): m/z 250 (MH^+, 25%).

Preparation 10 - 4-(1-t-Butoxycarbonylpiperidin-4-yl)pyrazole - 4-(1-t-Butoxycarbonyl-1,2,3,6-tetrahydropyridin-4-yl)pyrazole (0.35g) was dissolved in EtOH and hydrogenated over 10% Pd/C for 24 hours. Following filtration and concentration *in vacuo* the product was chromatographed (70% EtOAc/hexane) to give the title compound (0.26g).

Preparation 11 - 4-(1-Methyl-1,2,3,6-tetrahydropyridin-4-yl) pyrazole - A solution of 4-bromopyrazole (0.882g) in THF (15ml) at -70°C under argon was treated with a solution of n-BuLi (5.76ml of a 2.5M solution). The mixture was allowed to warm to room temperature, kept for 1½ hours and recooled to -70°C. 1-Methyl-4-piperidone (1.03ml) was added, the cooling bath removed and the mixture stirred 1½ hours, treated with acetic acid (5ml) and evaporated to dryness. The residue was dissolved in trifluoroacetic acid (10ml), heated to 70°C for 4 hours, evaporated to dryness. The residue was taken up in 40% aqueous potassium carbonate (10ml) and extracted 4 times with chloroform. The extracts were dried, evaporated and the residue chromatographed, eluting with chloroform/methanol/0.88NH_3(aq) (93:7:0.7) to give the title compound (0.52g). MS (NH_3 CI)m/z 164(MH^+, 80%)121(100%).

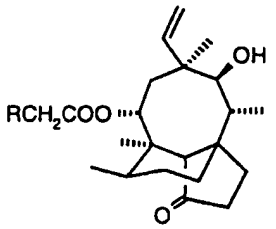
Preparation 12 - 4-(1-Methylpiperidin-4-yl)pyrazole - A solution of 4-(1-methyl-1,2,3,6-tetrahydropyridin-4-yl)pyrazole (0.25g) in ethanol (5ml) was treated with 2N HCl (1.5ml) and 10% Pd/C (100mg), stirred overnight under H_2 at atmospheric pressure, filtered through kieselguhr and evaporated. The residue was taken up in 40% aqueous K_2CO_3 , extracted 4 times with chloroform, the extracts dried and evaporated to give the title compound (0.25g). MS (+ve ion electrospray) m/z 166 (MH^+, 100%).

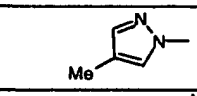
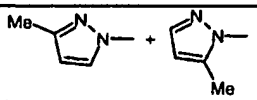
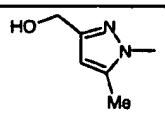
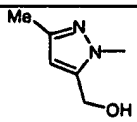
Preparation 13 – Benzyl 1,2,4-triazole-3-carboxylate - A suspension of 1,2,4-triazole-3-carboxylic acid (2.26g) in DMF (20ml) was treated with a 60% dispersion of NaH (0.8g), stirred overnight and treated with benzyl bromide (2.38ml). After stirring for 3 days, the mixture was diluted with EtOAc (100ml), washed with water (3 x 100ml), dried and evaporated to low volume. The precipitated solid was filtered off to give the title compound (0.43g). MS (APCI) m/z 202 ($M-H^-$, 100%).

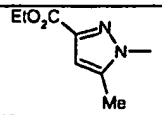
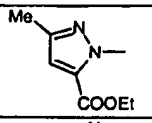
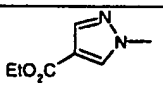
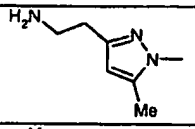
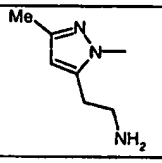
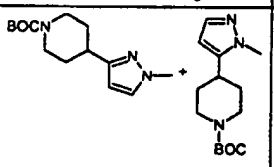
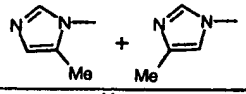
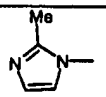
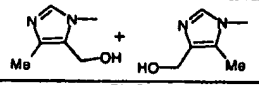
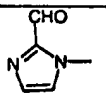
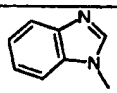
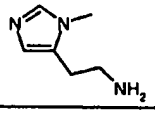
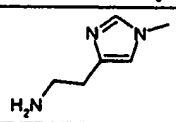
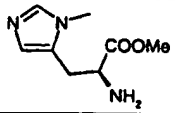
2-Dialkylaminomethylpyrroles were prepared according to W. Herz, K.Dittmer and S.J. Cristol, *J. Org. Chem.*, 1947, 69, 1698. **4-(1-Methylpiperidin-4-yl)imidazole** was prepared according to J.M. Arrang *et al.*, Eur.Pat.Appl. EP197840. **3-(Piperidin-4-yl)indole** was prepared according to D. Beck and K. Schenker, *Helv. Chim. Acta.*, 1968, 51, 260. **2-(Piperidin-4-yl)indole** was prepared according to World Patent WO 9747302. **3-(1-t-Butoxycarbonyl-1,2,3,6-tetrahydropyridin-4-yl)indole** was prepared according to J. Perregaard, K. Andersen, J. Hyttel and C. Sanchez, *J. Med. Chem.*, 1992, 35, 4813. **2-(Piperidin-4-yl)benzimidazole** was prepared according to C.G. Wahlgren and A.W. Addison, *J. Het. Chem.*, 1989, 26, 541

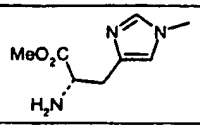
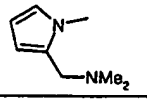
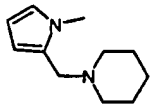
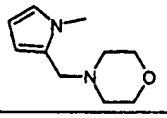
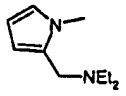
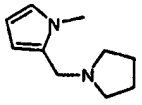
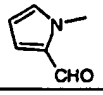
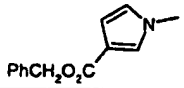
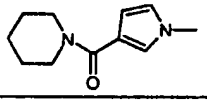
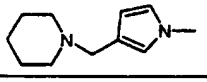
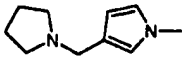
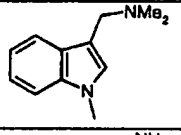
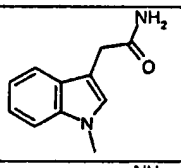
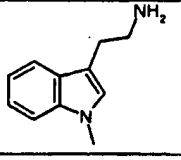
Example 1 - (4-Hydroxymethylpyrazole-1-yl)acetic acid mutilin 14-ester - A solution of 4-hydroxymethylpyrazole (0.29g) in DMF (19ml) was ice-cooled under argon and treated with sodium hydride (0.12g of a 60% dispersion in oil). After stirring for 1 hour, methanesulfonyloxyacetic acid mutilin 14-ester (1.37g, H. Egger and H. Reinshagen, *J. Antibiotics*, 1976, 29(9), 915) was added and stirring continued overnight at room temperature. The mixture was diluted with ethyl acetate (120ml), washed with water (3x100ml), dried ($MgSO_4$) and evaporated. The residue was chromatographed on silica, eluting with dichloromethane/methanol/0.88 NH_3 (aq)19:1:0.1 gave the title compound as a pale yellow foam (0.92g, 67%). MS (-ve ion electrospray): m/z 517, ($MOAc^-$, 100%) 457, ($M-H^-$,30%).

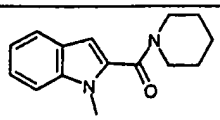
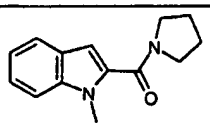
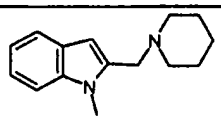
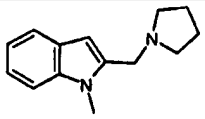
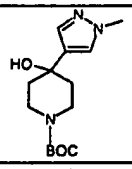
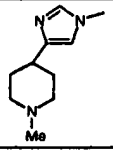
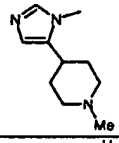
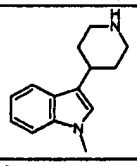
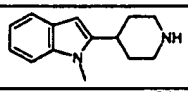
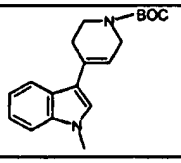
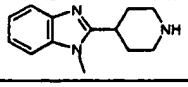
Example 2-48 were carried out likewise:

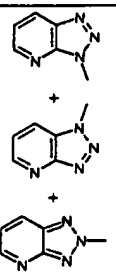
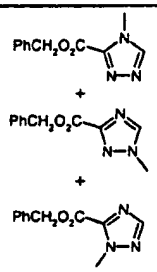
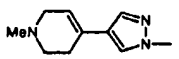
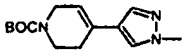


Example No.	R	Yield (%)	electrospray MS
2		52	(+ve ion) 885 ($2MH^+$, 90%), 443 (MH^+, 25%)
3		59	(+ve ion) 885 ($2MH^+$, 100%), 443 (MH^+, 80%)
4		25	(+ve ion) 473 (MH^+, 90%), 171 (100%)
5		22	(+ve ion) 473 (MH^+, 50%), 171 (100%)

6		39	(+ve ion) 515 (MH^+, 20%), 213 (100%)
7		16	(+ve ion) 515 (MH^+, 50%), 213 (100%)
8		42	(-ve ion) 499 ($M-H^-$, 100%)
9		34	(+ve ion) 486 (MH^+, 15%),184(100%)
10		9	(+ve ion) 486 (MH^+, 10%), 184 (100%)
11		77	
12		45	(+ve ion) 443 (MH^+, 100%)
13		73	(+ve ion) 443 (MH^+, 10%), 141 (100%)
14		26	(-ve ion) 531 ($MOAc^-$, 100%), 471 ($M-H^-$,80%)
15		58	(-ve ion) 515 ($MOAc^-$, 20%), 455 ($M-H^-$,100%)
16		42	(-ve ion) 530 ($MOAc^-$, 70%), 477 ($M-H^-$,100%)
17		9.5	(-ve ion) 530 ($MOAc^-$, 100%), 470 ($M-H^-$,80%)
18		4	(-ve ion) 530 ($MOAc^-$, 100%), 470 ($M-H^-$,90%)
19		5	(+ve ion) 530 (MH^+, 30%), 228 (100%)

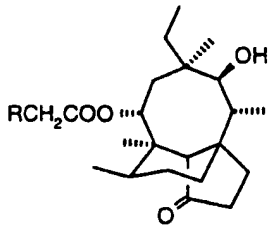
20		28	(+ve ion) 530 (MH^+, 50%), 228 (100%)
21		58	(-ve ion) 543 ($MOAc^-$, 100%), 483 ($M-H^-$, 15%)
22		16	(-ve ion) 583 ($MOAc^-$, 100%), 523 ($M-H^-$, 15%)
23		29	(-ve ion) 585 ($MOAc^-$, 100%), 525 ($M-H^-$, 40%)
24		15	(-ve ion) 571 ($MOAc^-$, 30%), 511 ($M-H^-$, 20%), 209 (100%)
25		11	(-ve ion) 569 ($MOAc^-$, 100%), 509 ($M-H^-$, 10%)
26		66	(-ve ion) 454 ($M-H^-$, 100%)
27		76	(-ve ion) 620 ($MOAc^-$, 90%), 560 ($M-H^-$,100%)
28		45	(+ve ion) 539 (MH^+, 100%)
29		13	(+ve ion) 525 (MH^+, 100%)
30		4	(+ve ion) 511 (MH^+, 100%), 440 (100%)*
31		43	(-ve ion) 593 ($MOAc^-$, 100%), 533 ($M-H^-$,25%)
32		33	(-ve ion) 593 ($MOAc^-$, 80%), 533 ($M-H^-$,100%)*
33		19	(+ve ion) 521 (MH^+, 100%)

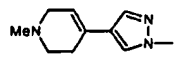
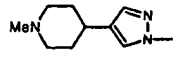
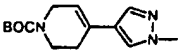
34		60	(-ve ion) 647 ($MOAc^-$, 100%). 587 ($M-H^-$.90%)
35		59	(-ve ion) 633 ($MOAc^-$, 100%), 573 ($M-H^-$.20%)
36		13	(+ve ion) 575 (MH^+, 40%). 273 (100%)
37		11	(+ve ion) 561 (MH^+, 30%). 188 (100%)
38		73	(+ve ion) 628 (MH^+, 100%)
39		15	(+ve ion) 526 (MH^+, 73%)
40		5	(+ve ion) 526 (MH^+, 28%)
41		37	(-ve ion) 559 ($M-H^-$, 60%)
42		31	(-ve ion) 559 ($M-H^-$, 100%)
43		52	(-ve ion APCI) 657 ($M-H^-$,40%)
44		47	(-ve ion) 560 ($M-H^-$ 100%)

45		86	(+ve ion) 481 (MH^+,100%)
46		46	(+ve ion) 564 (MH^+,100%)
47		90	(+ve ion) 524 (MH+, 40%), 179, 100%*
48		76	(+ve ion) 612 (MH+, 50%), 254 (100%)

*NMR Data - **Example 30** - ^1H NMR $\delta(CDCl_3)$(inter alia) 6.4-6.6(2H,m), 6.07(1H,t,J 3.1Hz), 5.95-6(1H,m), 5.77(1H,d,J 8.4Hz), 5.31(1H,dd,J 11 and 1.5 Hz), 5.18(1H,dd,J 17 and 1.5Hz), 4.75(2H,s), 3.52 and 3.43(2H, parts of Abq, J 13.5 Hz), 3.34 (1H, dd, J 11 and 6.7Hz). **Example 32** - ^1H NMR $\delta(CDCl_3)$(inter alia) 7.5-7.6(1H,m), 7.1-7.3(3H,m), 7.03(1H,s), 6.40(1H,dd, J 17.5 and 11Hz), 5.76(1H,d,J 8.4 Hz), 5.63(1H, broad s), 5.25-5.35(2H,m), 5.18(1H,dd,J 17.5 and 1.5 Hz), 4.75(2H,s), 3.73(2H,s), 3.32(1H,dd,J 10.5 and 6.3 Hz). **Example 47** - ^1H NMR $\delta(CDCl_3)$(inter alia) 7.58(1H,s), 7.34(1H,s), 6.44(1H,dd,J 17.5 and 10 Hz), 5.85-5.95(1H,m), 5.77(1H, d, J 8.5 Hz), 5.33(1H,dd,J 11 and 1.5 Hz), 5.19 (1H,dd,J 17 and 1.5 Hz), 4.77(1H,s), 3.34(1H,dd,J 11 and 6.8 Hz), 3.0-3.1(2H,m), 2.62(2H,t,J 5.5 Hz), 2.4-2.5(2H, m), 2.39(3H,s).

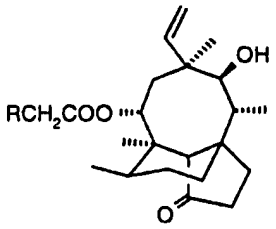
The following examples were carried out analogously to Example 1, using methanesulfonyloxyacetic acid 19,20-dihydro mutilin 14-ester instead of methanesulfonyloxyacetic acid mutilin 14-ester.

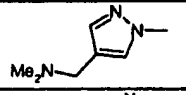
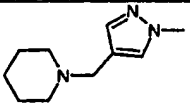
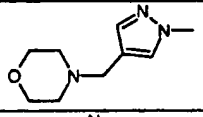
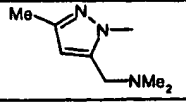


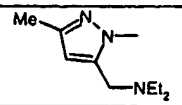
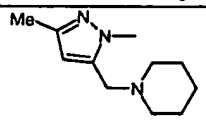
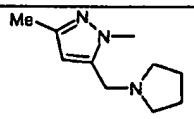
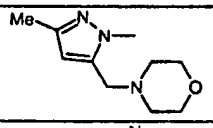
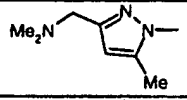
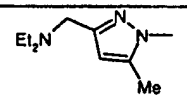
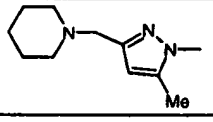
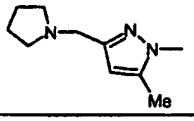
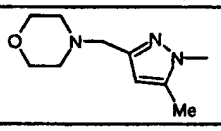
Example No.	R	Yield (%)	+ve ion electrospray MS
49		77	(+ve ion) 526 (MH^+, 50%), 179 (100%)
50		77	(+ve ion) 528 (MH^+, 100%)
51		76	(+ve ion) 612 (MH^+, 100%)

Example 52 - 4-Diethylaminomethylpyrazol-1-yl)acetic acid mutilin 14-ester - A solution of (4-hydroxymethylpyrazol-1-yl)acetic acid mutilin 14-ester (0.2g) in chloroform (3ml) was ice-cooled under argon, treated with triethylamine (0.116ml) and methanesulfonylchloride (0.064ml) and kept at room temperature overnight. Diethylamine (0.18ml) was added and the mixture stirred 24 hours, washed with water and aqueous $NaHCO_3$, dried ($MgSO_4$) and evaporated. Chromatography (dichloromethane/methanol/0.88NH_3 (aq)19:1:0.1) gave the title compound as a foam (0.1g,44%). MS (+ve ion electrospray): m/z 514, (MH^+,100%).

Examples 53 -65 were carried out likewise:

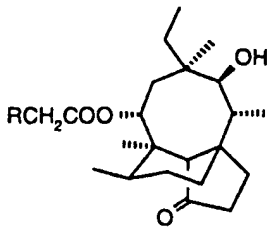


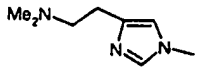
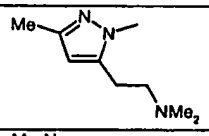
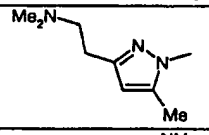
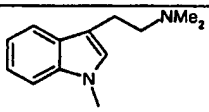
Example No.	R	Yield (%)	+ve ion electrospray MS
53		65	486 (MH^+,70%), 139 (100%)
54		17	526 (MH^+,100%)
55		72	528 (MH^+,100%)
56		28	500 (MH^+,20%), 198 (100%)

57		60	528 (MH^+,10%), 226 (100%)
58		75	540 (MH^+,10%), 238 (100%)
59		84	526 (MH^+,30%), 224 (100%)
60		47	542 (MH^+,20%), 240 (100%)
61		4	500 (MH^+,30%), 198 (100%)
62		4.5	528 (MH^+,50%), 226 (100%)
63		7.5	540 (MH^+,60%), 238 (100%)
64		84	526 (MH^+,80%), 224 (100%)
65		75	542 (MH^+,30%), 240 (100%)

Example 66 - [5-(2-Dimethylaminoethyl)imidazol-1-yl]acetic acid 19,20-dihydro-mutilin 14-ester - A solution of [5-(2-aminoethyl)imidazol-1-yl]acetic acid mutilin 14-ester (0.1g) in ethanol (5ml) was treated with formalin (0.15ml) and 10% Pd/C (0.2g) and stirred under H_2 at atmospheric pressure for 2 days. The mixture was filtered through kieselguhr and the filtrate evaporated to provide the title compound (0.066g, 62%). MS (+ve ion electrospray): m/z 502, (MH^+, 20%), 198 (100%).

Examples 67-70 were carried out likewise:



Example No.	R	Yield (%)	+ve ion electrospray MS
67		47	502 (MH^+,50%), 198 (100%)
68		55	516 (MH^+,40%), 212 (100%)
69		38	516 (MH^+,80%), 212 (100%)
70		31	551 (MH^+,30%), 247 (100%)

Example 71 - (5-Carboxy-3-methylpyrazol-1-yl)acetic acid mutilin 14-ester - A stirred solution of (5-ethoxycarbonyl-3-methylpyrazol-1-yl)acetic acid mutilin 14-ester (0.8g) in methanol (24ml) was treated dropwise over 15 minutes with a 0.5M aqueous solution of NaOH (3.42ml) and left overnight. Water (50ml) was added and the mixture washed with dichloromethane (50ml), acidified to pH3 with 1M HCl and extracted with dichloromethane (3x50ml). The combined extracts were dried ($MgSO_4$) and evaporated to give the title compound (0.46g, 61%). MS (-ve ion electrospray): m/z 485, ($M-H^-$, 100%).

Example 72 - (3-Carboxy-5-methylpyrazol-1-yl)acetic acid mutilin 14-ester - The title compound was produced analogously to example 71 (55%). MS (-ve ion electrospray): 485, ($M-H^-$, 100%).

Example 73 - (L-Histidin-1-yl) acetic acid mutilin 14-ester - Hydrolysis of (methyl L-histidin-1-yl)acetic acid mutilin 14 ester (example 19) was carried out analogously to example 71. However, the aqueous solution was acidified to pH 5, washed with dichloromethane and evaporated to dryness. The residue was triturated with ethanol, filtered and the filtrate evaporated to give the title compound (36%). MS (+ve ion electrospray): 516, (MH^+, 60%), 214(100%).

Example 74 - (L-Histidin-3-yl) acetic acid mutilin 14-ester - The title compound was produced analogously to example 73 (36%). MS (+ve ion electrospray) :m/z 516. (MH^+, 20%), 214, (100%).

Example 75 - (3-Carboxypyrrol-1-yl) acetic acid mutilin 14-ester - A solution of (3-benzyloxycarbonylpyrrol-1-yl)acetic acid mutilin ester (0.2g) in ethanol (5ml) was treated with 10% Pd/C (0.2g) and stirred under H_2 at atmospheric pressure overnight. The

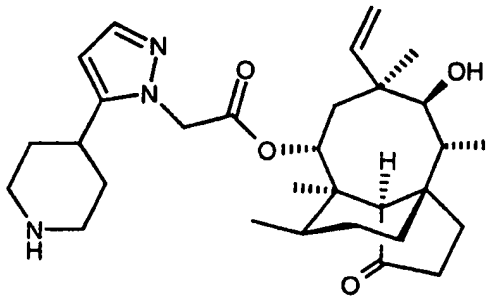
mixture was filtered through kieselguhr and the filtrate evaporated to yield the title compound (0.167g). MS (-ve ion electrospray): m/z 472, ($M-H^-$, 100%).

Example 76 - [3-(4-Methylpiperazin-1-yl)carbonyl-5-methylpyrazol-1-yl] acetic acid mutilin 14-ester - To an ice-cooled solution of (3-carboxy-5-methylpyrazol-l-yl)acetic acid mutilin 14-ester (0.2g) in dichloromethane (10ml) was added N-methylpiperazine (0.046ml) and then dicyclohexylcarbodiimide (0.13g). The mixture was allowed to reach room temperature, stirred overnight, filtered, washed with aqueous $NaHCO_3$, dried and evaporated. Chromatography provided the title compound as a foam (0.077g). MS (+ve ion electrospray): m/z 569, (MH^+, 50%), 267, (100%).

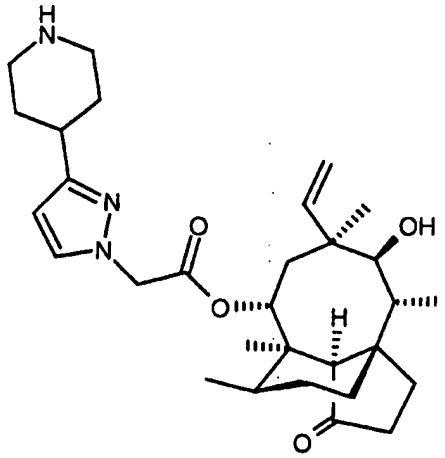
Example 77 - [3-N-(2-Dimethylaminoethyl)carboxamido-5-methylpyrazol-1-yl] acetic acid mutilin 14-ester - The title compound was produced in a similar manner to example 76 (11%). MS (+ve ion electrospray): m/z 557, (MH^+, 80%), 255, (80%), 210, (100%).

Example 78 - [5-N-(2-Dimethylaminoethyl)carboxamido-3-methylpyrazol-1-yl] acetic acid mutilin 14-ester - The title compound was produced in a similar manner to example 76 (12%). MS (+ve ion electrospray): m/z 557, (MH^+, 20%), 255, (100%), 210 (60%).

Example 79 - [5-(Piperidin-4-yl)pyrazol-1-yl]acetic acid mutilin 14-ester



and [3-(Piperidin-4-yl)pyrazol-1-yl]acetic acid mutilin 14-ester



A mixture of [5-(1-t-butoxycarbonylpiperidin-4-yl)pyrazol-1-yl]acetic acid mutilin 14-ester and [3-(1-t-butoxycarbonylpiperidin-4-yl)pyrazol-1-yl]acetic acid mutilin 14-ester (example 11, 4.1g) was dissolved in chloroform (20ml), treated with trifluoracetic acid (20ml) and left at room temperature for 4 hours. The mixture was evaporated to dryness, the residue partitioned between chloroform (50ml) and saturated aqueous $NaHCO_3$ (50ml) and the chloroform dried and evaporated. Chromatography (chloroform/methanol/35% NH_3 solution 9:1:0.1) separated the two title compounds. Less polar compound: [5-(piperidin-4-yl)pyrazol-1-yl] acetic acid mutilin 14-ester (1g), MS (+ve ion electrospray): m/z 512, (MH^+, 50%), 210, (100%), ^1H NMR $\delta(CDCl_3)$ (inter alia) 7.46(1H,d, J 1.8Hz), 6.43(1H,dd, J 17 and 11 Hz), 6.10(1H,d,J 1.8 Hz), 5.30(1H,dd,J 11 and 1Hz), 5.19(1H,dd,J 17 and 1 Hz), 4.84 and 4.74(2H, parts of ABq, J 17.5 Hz), 3.34(1H,d,J 6.1 Hz), 3.1-3.2(2H,m), 2.55-2.75(2H,m), 2.4-2.55(1H,m).

More polar compound: [3-(piperidin-4-yl)pyrazol-1-yl] acetic acid mutilin 14-ester (2.8g) MS (+ve ion electrospray): m/z 512, (MH^+, 90%), 210, (100%), ^1H NMR $\delta(CDCl_3)$ (inter alia) 7.32(1H,d, J 2.2 Hz), 6.45(1H,dd,J 17 and 11 Hz), 6.14(1H,d,J 2.2 Hz), 5.75(1H,d, J 8.5 Hz), 5.33(1H,d,J 11Hz), 5.19(1H,d,J 17Hz), 4.76(2H,s), 3.2-3.4(3H,m), 2.7-2.9(3H,m).

Example 80 – [3-(1,2,3,6-Tetrahydropyridin-4-yl)indol-1-yl]acetic acid mutilin 14-ester - The title compound was prepared from [3-(1-t-butoxycarbonyl-1,2,3,6-tetrahydropyridin-4-yl)indol-1-yl]acetic acid mutilin 14-ester according to the procedure of Example 79 (47%). MS (-ve ion electrospray) m/z 557 ($M-H^-$, 80%).

Example 81 – [4-(1,2,3,6-Tetrahydropyridin-4-yl)pyrazol-1-yl]acetic acid mutilin 14-ester - The title compound was prepared from [4-(1-t-butoxycarbonyl-4-hydroxypiperidin-4-yl)pyrazol-1-yl]acetic acid mutilin 14-ester according to the procedure of Example 79 (68%). MS (+ve ion electrospray) m/z 510 (MH^+, 100%).

Example 82 - [4-(Piperidin-4-yl)pyrazol-1-yl]acetic acid mutilin 14-ester - The title compound was prepared from [4-(1-t-butoxy carbonylpiperidin-4-yl)pyrazol-1-yl]acetic acid mutilin 14-ester according to the procedure of example 79 (0.318g) MS (+ve ion electrospray):m/z 512 (MH^+, 100%).

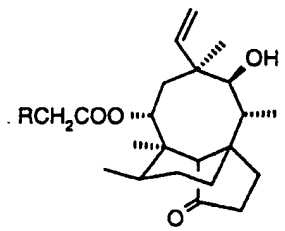
Example 83 - [4-(1,2,3,6-Tetrahydropyridin-4-yl)pyrazol-1-yl]acetic acid 19,20-dihydromutilin 14-ester - The title compound was prepared from [4-(1-butoxycarbonyl 1,2,3,6-tetrahydropyridin-4-yl)pyrazol-1-yl]acetic acid 19,20-dihydromutilin 14-ester according to the procedure of example 79 (0.366g. MS (-ve ion electrospray):m/z 510 ($M-H^-$100%).

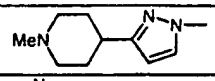
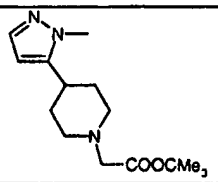
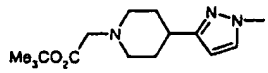
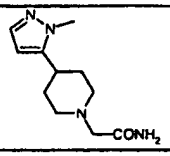
Example 84 - [4-(1-Methylpiperidin-4-yl)pyrazol-1-yl]acetic acid mutilin 14-ester-11-formate - A solution of [4-(1,2,3,6 tetrahydropyridin-4-yl)pyrazol-1-yl]acetic acid mutilin 14-ester (0.4g) in 98% formic acid (2ml) was treated dropwise with 37% aqueous formaldehyde (1ml) over 15 minutes and then refluxed 18 hours. After evaporation to dryness, the residue was taken up in water (10ml)/EtOAc (10ml) and the aqueous pH adjusted to 9-10 with dilute NaOH. The organic was separated, dried and evaporated. Chromatography (dichloromethane/methanol/0.88 NH_3(aq) 9:1:0.1) gave the title compound.

Example 85 - [4-(1-Methylpiperidin-4-yl)pyrazol-1-yl]acetic acid mutilin 14-ester - A solution of [4-(methylpiperidin-4-yl)pyrazol-1-yl]acetic acid mutilin 14-ester-11-formate (0.1g) in ethanol was treated dropwise with 0.5N ethanolic KOH (0.36ml), left 1 hour and partitioned between water (30ml) and EtOAc (30ml). The organic layer was dried and evaporated and the residue chromatographed (dichloromethane/methanol/0.88NH_3 (aq) 98:2:0.2 then 95:5:0.5 then 90:10:1) to give the title compound.

Example 86 - [5-(1-Methylpiperidin-4-yl)pyrazol-1-yl] acetic acid mutilin 14-ester - A solution of [5-(piperidin-4-yl)pyrazol-1-yl] acetic acid mutilin 14-ester (0.11g) in acetonitrile (4ml) was treated with K_2CO_3 (0.07g) and MeI (0.016ml), stirred overnight, filtered through kieselguhr and evaporated. Chromatography (chloroform/methanol/35% NH_3 solution 19:1:0.1) provided the title compound (0.046g). MS (+ve ion electrospray): m/z 526, (MH^+, 80%), 224, (100%).

Example 87-90 were carried out likewise:

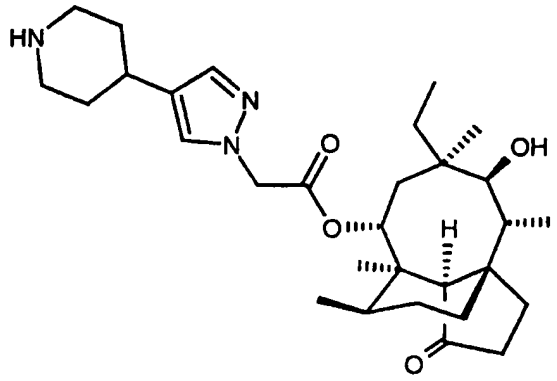


Example No.	R	Yield (%)	+ve ion electrospray MS
87		32	526 (MH^+, 100%)
88		98	626 (MH^+,70%) 570 (100%)
89		81	626 MH^+,95%) 570 (100%)
90		97	569 (MH^+, 20%) 267 (100%)

Example 91 - [5-(1-Carboxymethylpiperidin-4-yl)pyrazol-1-yl] acetic acid mutilin 14-ester - A solution of [5-(1-t-butoxycarbonylpiperidin-4-yl)pyrazol-1-yl]acetic acid mutilin 14-ester (0.1g) in chloroform (2ml)/trifluoroacetic acid (3ml) was kept 3 hours and evaporated. Chromatography(dichloromethane/methanol/35% NH_3 solution 4:1:0.1) provided the title compound as a foam (0.9g). MS (-ve ion electrospray): m/z 568, ($M-H^-$, 100%).

Example 92 - [3-(1-Carboxymethylpiperidin-4-yl)pyrazol-1-yl] acetic acid mutilin 14-ester - The title compound was prepared as for example 91 (99%). MS (-ve ion electrospray) 568 ($M-H^-$,100%).

Example 93 – [4-(Piperidin-4-yl)pyrazol-1-yl]acetic acid 19, 20-dihydromutilin 14-ester



A solution of [4-(1,2,3,6-tetrahydropyridin-4-yl)pyrazol-1-yl]acetic acid mutilin 14-ester (0.4g) in ethanol (20ml) was treated with 10% Pd/C (0.1g) and shaken under hydrogen at 50 psi for 18 hours. Catalyst was filtered off and the filtrate evaporated to give the title

compound (0.36g). MS (+ve ion electrospray) m/z 514 (MH^+, 50%), ^1H NMR $\delta(CDCl_3)$ (inter alia) 7.39(1H,s), 7.21(1H,s), 5.64(1H,d,J 8 Hz), 4.77(2H,s), 3.39(1H,d,J 6.0 Hz), 3.20(2H,d,J 12.4 Hz), 2.7-2.8(2H,m), 2.6-2.7(1H,m).

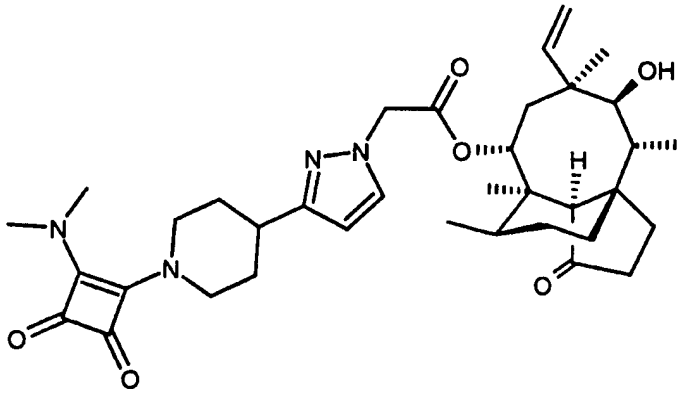
Example 94 – {5-[1-(1,2-Dioxo-3-ethoxy-3-cyclobuten-4-yl)piperidin-4-yl]pyrazol-1-yl}acetic acid mutilin 14-ester - A solution of [5-(piperidin-4-yl)pyrazol-1-yl]acetic acid mutilin 14-ester (0.13g) in ethanol (2ml) was treated with 3,4-diethoxy-3-cyclobutene-1,2-dione (0.1ml). After 4 days the solid was filtered off to give the title compound (0.16g). MS (-ve ion electrospray) m/z 694 ($MOAc^-$, 100%), 634 ($M-H^-$, 70%).

Example 95 – {3-[1-(1,2-Dioxo-3-ethoxy-3-cyclobuten-4-yl)piperidin-4-yl]pyrazol-1-yl}acetic acid mutilin 14-ester - The title compound was prepared according to the procedure of Example 94 (100%).

Example 96 – {5-[1-(1,2-Dioxo-3-amino-3-cyclobuten-4-yl)piperidin-4-yl]pyrazol-1-yl}acetic acid mutilin 14-ester - A solution of {5-[1-(1,2-dioxo-3-ethoxy-3-cyclobuten-4-yl)piperidin-4-yl]pyrazol-1-yl}acetic acid mutilin 14-ester (0.14g) in 2M ethanolic ammonia was kept 4 hours and evaporated. The residue was taken up in chloroform, filtered and the filtrate applied to a column. Elution with chloroform/methanol/0.88NH_3(aq) 9:1:0.1 provided the title compound (108mg). MS(-ve ion electrospray) m/z 605 ($M-H^-$, 100%).

Example 97 – {3-[1-(1,2-Dioxo-3-amino-3-cyclobuten-4-yl)piperidin-4-yl]pyrazol-1-yl}acetic acid mutilin 14-ester - The title compound was prepared according to the procedure of Example 96 (88%). MS (-ve ion electrospray) m/z 605 ($M-H^-$, 100%).

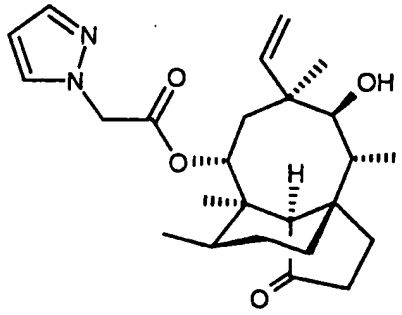
Example 98 – {3-[1-(1,2-Dioxo-3-dimethylamino-3-cyclobuten-4-yl)piperidin-4-yl]pyrazol-1-yl}acetic acid mutilin 14-ester



The title compound was prepared analogously to the procedure of Example 96, using a solution of approximately 2M dimethylamine in ethanol (36%). MS (-ve ion electrospray) m/z 693 ($MOAc^-$, 80%) 633 ($M-H^-$, 100%), ^1H NMR $\delta(CDCl_3)$(inter alia)

7.34(1H,d,J 2.5 Hz), 6.45(1H, dd, J 17 and 11 Hz), 6.13(1H,d,J 2.5 Hz), 5.75(1H,d,J 8.5 Hz), 5.33(1H,dd, J 11 and 1.3Hz), 5.20 (1H,dd,J 17 and 1.3Hz), 4.76(2H,s), 4.1-4.25(2H,m), 3.2-3.4(3H,m), 3.23(6H,s), 2.8-3.0(1H,m).

Example 99 – (Pyrazol-1-yl)acetic acid mutilin 14-ester



A solution of 4-bromopyrazole (2g) in THF (30ml) at -78°C was treated with n-BuLi (19ml of a 1.6M solution), allowed to warm to room temp. and kept 1.5 hours. The mixture was cooled to -78°C and a 25ml aliquot was added dropwise to a solution of methanesulfonyloxyacetic acid mutilin 14-ester (3.13g) in THF (30ml). The mixture was warmed to room temp., kept 16 hours and evaporated. Chromatography (EtOAc/hexane 2:3) gave the title compound (0.2g) MS (+ve ion electrospray m/z 429 (MH^+, 100%), ^1H NMR $\delta(CDCl_3)$(inter alia) 7.54(1H,d,J 1.6Hz), 7.43(1H,d,J 2.2 Hz), 6.45(1H,dd,J 17 and 11 Hz), 6.33(1H,t, J 2.2 and 1.6Hz), 5.78(1H,d, J 8.5 Hz), 5.33(1H,d,J 11Hz), 5.20(1H,d,J 17Hz), 4.84(2H,s), 3.34(1H,dd J 10.5 and 6.5Hz).

Example 100 - (Tetrazol-2-yl)acetic acid mutilin 14-ester - A solution of pleuromutilin (0.376g), tetrazole (0.105g) and triphenylphosphine (0.314g) in THF (10ml) was cooled to -10°C and treated with diethyl azodicarboxylate (0.19ml) and allowed to warm to room temperature. After 2 hours the solvent was evaporated and the residue chromatographed (EtOAc/hexane mixtures) to give the title compound (0.23g). MS (+ve ion electrospray): m/z 431, (MH^+,10%), 279(100%).

Example 101 - (5-Phenyltetrazol-2-yl) acetic acid mutilin 14-ester- The title compound was obtained by the same procedure as example 100 (40%). MS (+ve ion electrospray): m/z 507, (MH^+, 100%)

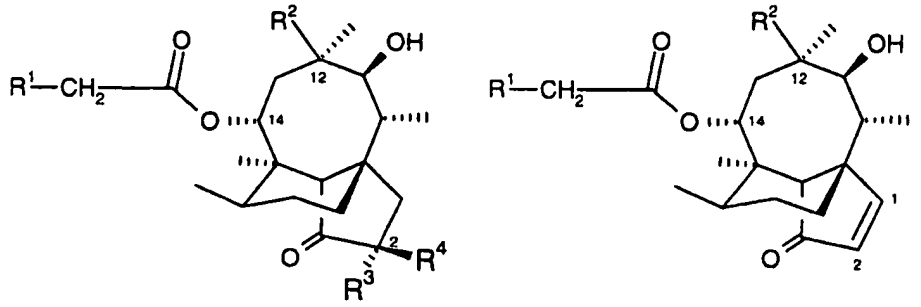
Biological Data

Compounds of the present invention were assessed for anti-bacterial activity in a conventional MIC assay against a range of pathogenic organisms.

Compounds were found to have MICs in the range 0.06 to 16 μg/ml against *Staph Aureus* Oxford and 0.06 to 16 μg/ml against *Strep Pneumoniae* (r6).

Claims

1. A compound of formula (IA) or (IB):



(IA) (IB)

in which:

R^1 is an optionally substituted heteroaryl group which comprises a five membered heteroaromatic ring which has at least one nitrogen atom and which is linked via a nitrogen atom;

R^2 is vinyl or ethyl;

R^3 is H, OH or F, and R^4 is H, or R^3 is H and R^4.

2. A compound according to claim 1 in which R^1 is pyrrole, pyrazole, imidazole, 1,2,3-triazole, 1,2,4-triazole, tetrazole, indole, benzimidazole, benzotriazole, 2-aza-indole or 6-aza-indole.

3. A compound according to claim 1 or claim 2 in which R^1 is pyrrole or pyrazole.

4. A compound according to any one of claims 1 to 3 in which R^1 is substituted by mono- or di-(C1-6)alkylamino, mono- or di-(C_{1-6})alkylaminoalkyl, N-heterocyclyl or N-heterocyclo-(C_{1-6})alkyl.

5. A compound according to any one of claims 1 to 4 in which R^1 is substituted by 2-aminoethyl, 2-dimethylaminoethyl, piperidinyl, piperidinomethyl, morpholinomethyl or pyrrolidinomethyl.

6. A compound according to claim 1 selected from:

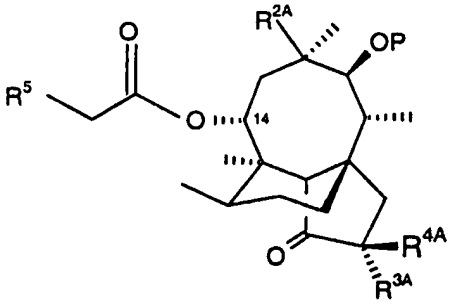
[3-(Pyrrolidin-1-ylmethyl)pyrrol-1-yl]acetic acid mutilin 14-ester;

[3-(Carboxamidomethyl)indol-1-yl]acetic acid mutilin 14-ester;

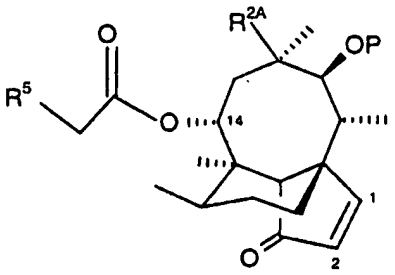
[3-(2-Aminoethyl)indol-1-yl]acetic acid mutilin 14-ester;

[4-(1-Methyl-1,2,3,6-tetrahydropyridin-4-yl)pyrazol-1-yl]acetic acid mutilin 14-ester;
[5-(Piperidin-4-yl)pyrazol-1-yl]acetic acid mutilin 14-ester;
[3-(Piperidin-4-yl)pyrazol-1-yl]acetic acid mutilin 14-ester;
[4-(Piperidin-4-yl)pyrazol-1-yl]acetic acid 19,20-dihydro-mutilin 14-ester;
{3-[1-(1,2-Dioxo–3-dimethylamino-3-cyclobuten-4-yl)piperidin-4-yl]pyrazol-1-yl}acetic acid mutilin 14-ester; and
(Pyrazol-1-yl)acetic acid mutilin 14-ester.

7. A method of preparing a compound of formula (IA) or (IB) according to any one of claims 1 to 6 which comprises reacting a compound of formula (IIA) or (IIB):



(IIA)



(IIB)

in which:
R^5 is a leaving group or hydroxy, P is hydrogen or a removable hydroxy-protecting group, and R^{2A}, R^{3A} and R^{4A} are R^2, R^3 and R^4 as defined for formulae (IA) and (IB) or groups convertible to R^2, R^3 and R^4;
(i) when R^5 is a leaving group, with R^1H under alkylating conditions; or
(ii) when R^5 is a hydroxy, with $R^{1A}H$ in which R^{1A} is a group R^1 which contains an acidic NH, under Mitsunobu conditions;
and thereafter, and if so needed;
converting P to hydrogen,
converting an R^{2A}, R^{3A} or R^{4A} group to an R^2, R^3 or R^4 group, and/or
converting one R^1, R^2, R^3or R^4 group to another R^1, R^2, R^3or R^4 group.

8. A pharmaceutical composition comprising a compound according to any one of claims 1 to 6, or a compound obtainable by a process according to claim 7, or a pharmaceutically acceptable salt or derivative thereof, and a pharmaceutically acceptable carrier.

9. A method of treating microbial infections in animals, especially in humans and in domesticated mammals, which comprises administering a compound according to any one of claims 1 to 6, or a compound obtainable by a process according to claim 7, or a pharmaceutically acceptable salt or derivative thereof, or a composition according to the invention, to a patient in need thereof.

10. A method of treating or preventing recurrent otitis media or recurrent acute bacterial sinusitis in humans, which comprises nasally administering a compound according to any one of claims 1 to 6, or a compound obtainable by a process according to claim 7, or a pharmaceutically acceptable salt or derivative thereof, or a composition according to the invention, to a patient in need thereof.

11. A method of treatment of skin and soft tissue infections and in the treatment of acne in humans, which comprises topically administering a compound according to any one of claims 1 to 6, or a compound obtainable by a process according to claim 7, or a pharmaceutically acceptable salt or derivative thereof, or a composition according to the invention, to a patient in need thereof.

INTERNATIONAL SEARCH REPORT

Inte Jonal Application No
PCT/EP 99/09577

A. CLASSIFICATION OF SUBJECT MATTER
IPC 7 A61K31/415 A61K31/445 A61K31/40 A61K31/405 C07D231/12
C07D231/14 C07D209/14 C07D209/18 C07D207/32 C07D207/40
C07D209/42 A61P31/04 C07D401/04 C07D231/56 C07D233/54

According to International Patent Classification (IPC) or to both national classification and IPC

B. FIELDS SEARCHED

Minimum documentation searched (classification system followed by classification symbols)
IPC 7 C07D A61K

Documentation searched other than minimum documentation to the extent that such documents are included in the fields searched

Electronic data base consulted during the international search (name of data base and, where practical, search terms used)

C. DOCUMENTS CONSIDERED TO BE RELEVANT

Category *	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
A	US 4 278 674 A (REINSHAGEN HELLMUTH ET AL) 14 July 1981 (1981-07-14) the whole document	1-11
A	WO 97 25309 A (HUNT ERIC ;HINKS JEREMY DAVID (GB); SMITHKLINE BEECHAM PLC (GB); T) 17 July 1997 (1997-07-17) cited in the application claims	1-11
A	WO 98 05659 A (NAYLOR ANTOINETTE ;HUNT ERIC (GB); SMITHKLINE BEECHAM PLC (GB); TA) 12 February 1998 (1998-02-12) cited in the application claims	1-11

☐ Further documents are listed in the continuation of box C.

☒ Patent family members are listed in annex.

* Special categories of cited documents :

"A" document defining the general state of the art which is not considered to be of particular relevance

"E" earlier document but published on or after the international filing date

"L" document which may throw doubts on priority claim(s) or which is cited to establish the publication date of another citation or other special reason (as specified)

"O" document referring to an oral disclosure, use, exhibition or other means

"P" document published prior to the international filing date but later than the priority date claimed

"T" later document published after the international filing date or priority date and not in conflict with the application but cited to understand the principle or theory underlying the invention

"X" document of particular relevance; the claimed invention cannot be considered novel or cannot be considered to involve an inventive step when the document is taken alone

"Y" document of particular relevance; the claimed invention cannot be considered to involve an inventive step when the document is combined with one or more other such documents, such combination being obvious to a person skilled in the art.

"&" document member of the same patent family

Date of the actual completion of the international search	Date of mailing of the international search report
19 May 2000	06/06/2000

Name and mailing address of the ISA	Authorized officer
European Patent Office, P.B. 5818 Patentlaan 2 NL - 2280 HV Rijswijk Tel. (+31-70) 340-2040, Tx. 31 651 epo nl, Fax: (+31-70) 340-3016	Chouly, J

1

Form PCT/ISA/210 (second sheet) (July 1992)

INTERNATIONAL SEARCH REPORT

Int. Jonal Application No
PCT/EP 99/09577

A. CLASSIFICATION OF SUBJECT MATTER
IPC 7 C07D471/04 C07D249/10 //(C07D471/04,249:00,221:00)

According to International Patent Classification (IPC) or to both national classification and IPC

B. FIELDS SEARCHED

Minimum documentation searched (classification system followed by classification symbols)

Documentation searched other than minimum documentation to the extent that such documents are included in the fields searched

Electronic data base consulted during the international search (name of data base and, where practical, search terms used)

C. DOCUMENTS CONSIDERED TO BE RELEVANT

Category *	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.

☐ Further documents are listed in the continuation of box C.

☒ Patent family members are listed in annex.

* Special categories of cited documents :

"A" document defining the general state of the art which is not considered to be of particular relevance

"E" earlier document but published on or after the international filing date

"L" document which may throw doubts on priority claim(s) or which is cited to establish the publication date of another citation or other special reason (as specified)

"O" document referring to an oral disclosure, use, exhibition or other means

"P" document published prior to the international filing date but later than the priority date claimed

"T" later document published after the international filing date or priority date and not in conflict with the application but cited to understand the principle or theory underlying the invention

"X" document of particular relevance; the claimed invention cannot be considered novel or cannot be considered to involve an inventive step when the document is taken alone

"Y" document of particular relevance; the claimed invention cannot be considered to involve an inventive step when the document is combined with one or more other such documents, such combination being obvious to a person skilled in the art.

"&" document member of the same patent family

Date of the actual completion of the international search	Date of mailing of the international search report
19 May 2000	

Name and mailing address of the ISA	Authorized officer
European Patent Office, P.B. 5818 Patentlaan 2 NL – 2280 HV Rijswijk Tel. (+31-70) 340-2040, Tx. 31 651 epo nl, Fax: (+31-70) 340-3016	Chouly, J

1

Form PCT/ISA/210 (second sheet) (July 1992)

INTERNATIONAL SEARCH REPORT

Information on patent family members

Int. Ional Application No
PCT/EP 99/09577

Patent document cited in search report	Publication date	Patent family member(s)	Publication date
US 4278674 A	14-07-1981	CH 572893 A	27-02-1976
		CH 568272 A	31-10-1975
		CH 575375 A	14-05-1976
		CH 560180 A	27-03-1975
		CH 559719 A	14-03-1975
		AT 335054 B	25-02-1977
		AT 845872 A	15-06-1976
		BE 789629 A	03-04-1973
		BG 21386 A	20-05-1976
		BG 20773 A	20-12-1975
		CA 979447 A	09-12-1975
		CS 219306 B	25-03-1983
		CY 987 A	02-08-1979
		DD 100706 A	05-10-1973
		DE 2248237 A	12-04-1973
		DE 2265666 C	03-02-1983
		DK 139720 B	02-04-1979
		DK 498076 A,B,	03-11-1976
		DK 498176 A,B,	03-11-1976
		FI 58635 B	28-11-1980
		FI 794060 A,B,	27-12-1979
		FI 794061 A,B,	27-12-1979
		FR 2157828 A	08-06-1973
		GB 1410506 A	15-10-1975
		GB 1410505 A	15-10-1975
		HK 25379 A	27-04-1979
		HU 168436 B	28-04-1976
		IE 37699 B	28-09-1977
		IE 37700 B	28-09-1977
		IL 40494 A	30-01-1976
		IL 47262 A	30-01-1976
		JP 48044244 A	26-06-1973
		JP 57007147 B	09-02-1982
		MY 10379 A	31-12-1979
		NL 7213254 A,B,	09-04-1973
		NO 136459 B	31-05-1977
		NO 136837 B	08-08-1977
		NO 137440 B	21-11-1977
		OA 4192 A	31-12-1979
		PH 11870 A	04-08-1978
		RO 67478 A	15-03-1980
		RO 67479 A	21-06-1981
		SE 398348 B	19-12-1977
		SE 424074 B	28-06-1982
		SE 7509456 A	26-08-1975
		SE 419984 B	07-09-1981
		SE 7509457 A	26-08-1975
		US 4107164 A	15-08-1978
		US 4208326 A	17-06-1980
		YU 249772 A	27-04-1983
WO 9725309 A	17-07-1997	AU 715229 B	20-01-2000
		AU 1307897 A	01-08-1997
		BG 102600 A	30-09-1999
		BR 9612426 A	13-07-1999
		CA 2240467 A	17-07-1997
		CN 1214039 A	14-04-1999
		CZ 9802124 A	16-12-1998

INTERNATIONAL SEARCH REPORT

Information on patent family members

Int. Jonal Application No
PCT/EP 99/09577

Patent document cited in search report	Publication date	Patent family member(s)	Publication date
WO 9725309 A		EP 0874809 A	04-11-1998
		HU 9900973 A	30-08-1999
		NO 983074 A	31-08-1998
		PL 327737 A	21-12-1998
		SK 91098 A	11-02-1999
		US 6020368 A	01-02-2000
		ZA 9700017 A	02-07-1998
		AU 4203697 A	25-02-1998
		CN 1231665 A	13-10-1999
		CZ 9900293 A	16-06-1999
		WO 9805659 A	12-02-1998
		EP 0934316 A	11-08-1999
		NO 990463 A	01-02-1999
		PL 331470 A	19-07-1999
		ZA 9706817 A	01-02-1999
WO 9805659 A	12-02-1998	AU 715229 B	20-01-2000
		AU 1307897 A	01-08-1997
		AU 4203697 A	25-02-1998
		BG 102600 A	30-09-1999
		BR 9612426 A	13-07-1999
		CN 1231665 A	13-10-1999
		CZ 9900293 A	16-06-1999
		EP 0874809 A	04-11-1998
		EP 0934316 A	11-08-1999
		NO 983074 A	31-08-1998
		NO 990463 A	01-02-1999
		PL 327737 A	21-12-1998
		PL 331470 A	19-07-1999
		SK 91098 A	11-02-1999
		US 6020368 A	01-02-2000
		CA 2240467 A	17-07-1997
		CN 1214039 A	14-04-1999
		CZ 9802124 A	16-12-1998
		WO 9725309 A	17-07-1997
		HU 9900973 A	30-08-1999
		ZA 9706817 A	01-02-1999